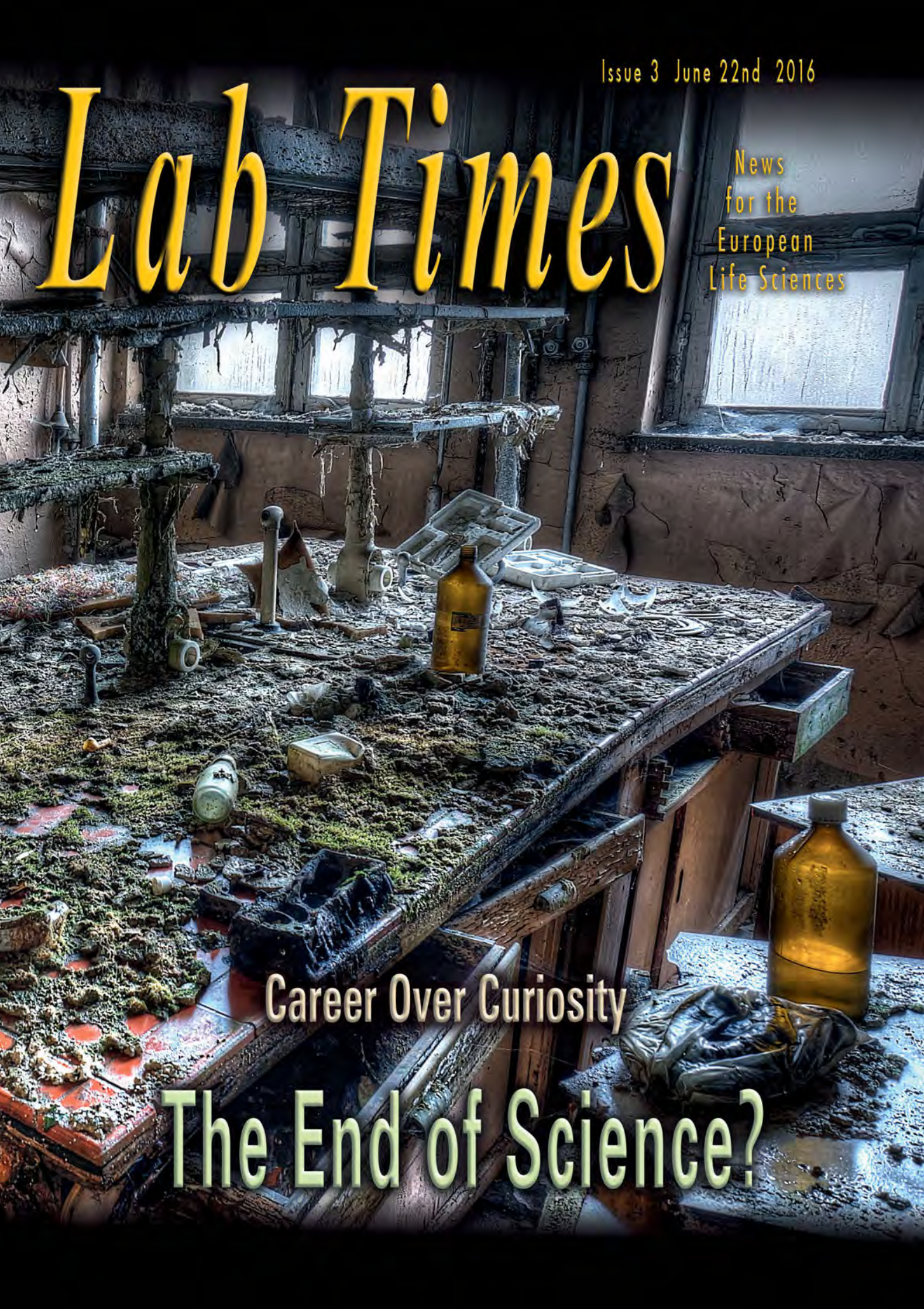
Issue 3 June 22nd 2016

Lab Times

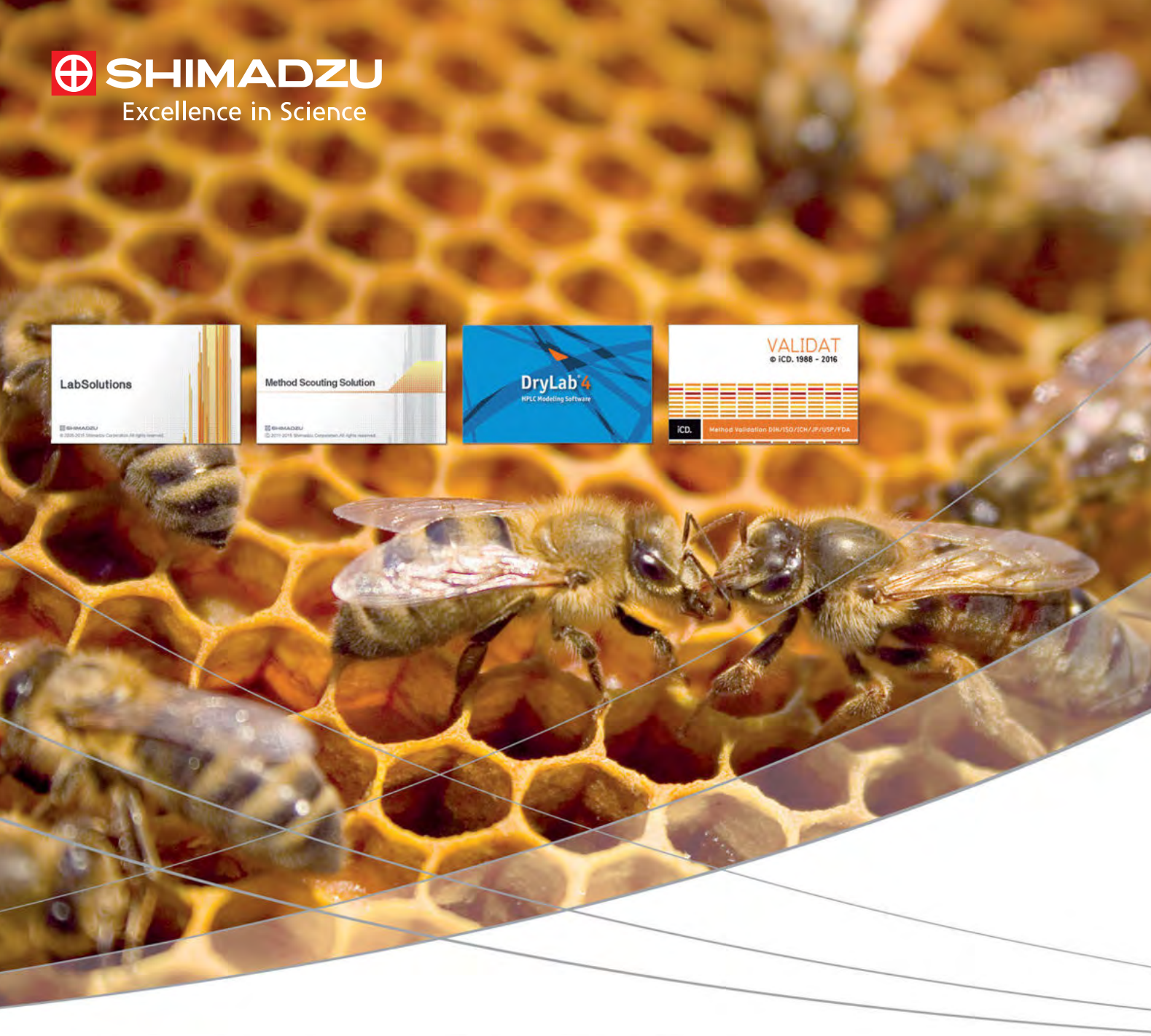
News for the European Life Sciences



Career Over Curiosity

The End of Science?

SHIMADZU
Excellence in Science



Clever co-workers

Outstanding platform for automated method development

The Nexera Method Scouting system provides an all-round solution for efficient HPLC method development and implementation. The automated method development solution comprises of four software packages which complement each other in creating a seamless method development work-flow.

- **Method Scouting Solution software**
  enables automated, quick and simple column and solvent screening
- **LabSolutions software**
  for data evaluation
- **DryLab®4 HPLC modeling software**
  focuses on strategic method optimization by calculation and visualization of the design space
- **VALIDAT®**
  for an automated method validation workflow from preliminary plan to a fully customized validation report

www.shimadzu.eu

"A Reliable Scientific Literature...

...is crucial for an efficient research process," wrote Bernd Pulverer, EMBO's Head of Scientific Publication, in an editorial for *The EMBO Journal* last year (34(20):2483-5). An increasing number of retractions shows, however, that we cannot always trust on what's written. In recent times, the term "correcting the scientific record" has become somewhat of a recurring catchphrase.

The world of academic publishing, currently, has three options how to handle papers with wrong, faulty or misleading findings. Depending on the kind and severity of the misinformation, journals can either issue an erratum, a correction, an expression of concern and, as the last and final step, a retraction.

COPE, the Committee of Publication Ethics, advises that journal editors should only then consider retracting a study when "they have clear evidence that the findings are unreliable, either as a result of misconduct (e.g. data fabrication) or honest error (e.g. miscalculation or experimental error); the findings have previously been published elsewhere without proper cross-referencing, permission or justification (i.e. cases of redundant publication); it constitutes plagiarism or it reports unethical research."

Photo: designerspics.com/Jeshu John

The guidelines are clear about it but is it always the right decision to retract a study when published data is found to be erroneous?

Hanna Kokko, evolutionary biologist at the University of Zürich, recently set a precedent and, with it, ignited a lively debate on social media and *Retraction Watch*. What had happened?

When working on a new paper about mate searching, Kokko's new collaborator, Lutz Fromhage from the University of Jyväskylä, Finland, noticed an analytical mistake in one of her earlier papers. The two worked it out and wrote a new paper, correcting the mistake and updating their conclusions. The journal editors of *Evolution*, where the original study had been published and the new manuscript submitted, now had to decide what to do: add an erratum to the out-dated study, explaining the mistake and linking to the new study, or erase it from the scientific record. They went for the latter option, also because an erratum can only be issued when it is signed by the same authors as the original study. Which wasn't possible in this case. Hence, study retracted! Self-criticism felled a paper. Bam!

Although a few scientists agreed with the journal, most were outraged at this editorial decision: open science advocate, Mike Taylor from the University of Bristol, for instance, commented, "The journal is wrong, simply and flatly. I hope they have a decency and honesty to undo their error, and unretract the original paper. If they don't, then they are sending a simple message: 'Never admit to any weaknesses in your own earlier work, or it might get retracted; and invest time pointing out weaknesses in your rivals' earlier work, in the hope that yours will look better in comparison when their work is retracted.' It's hard to imagine a message more inimical to the advance of science." Daniele Fanelli, well-known for his studies on scientific misconduct and research bias, added, "This journal's actions reflect a complete misunderstanding of what retractions should be used for, and they set a damaging precedent that will discourage open debate, self-criticism and self correction."

Also, Kokko thinks that researchers, who notice mistakes in their earlier work, might now keep quiet for fear of retraction. "In our view, science is best served if (a) papers identifying a scientific problem and making initial attempts at solving them are, generally, kept in the literature even if flaws are later found, as there is usually quite a bit of merit left in the sense of identifying areas of research; (b) if I was criticising someone else's work (other than my own) at a similar level of severity, I would still not want that person to retract (or to be forced to retract) the original, for that would leave counterproductive gaps in literature and change research atmospheres towards a much more destructive game," she told *Retraction Watch*.

Ben Ashby, who was one of the current paper's referees, even says that an erratum had achieved the same thing as a retraction, without stigmatising the authors. In his reviewer report, he had thus suggested a correction. "Scientists are not perfect. Honest mistakes will happen along the way, but it's important that we encourage people to admit when they have gone wrong," he writes in a guest post for *Retraction Watch*.

Was it now right or wrong to retract Hanna Kokko's erroneous study? Opinions will differ. But, when considering this case, it's clear that journal editors have a responsibility beyond "merely" correcting the scientific record. With their decisions, they can, at the very least, push science towards more honesty and openness or, *vice versa*, towards dishonesty and irresponsibilty.

Photo: HPI

With the help of designer recombinases, virologist Joachim Hauber was able to reverse an HIV infection at the molecular level. Now, he's ready to take the next big step. (p. 24)

Photo: Lida Xing, Univ Beijing

You don't want to be trapped under a giant sauropod's foot. Karl Bates and Vivian Allen used computer power to reconstruct the evolution of the ancient titans' body plan. (p. 32)

Photo: Rodos BT

The most effective medicine cannot work if it doesn't reach its destination. Or, even worse, if it works in the wrong place. These three men might be able to help. (p. 39)

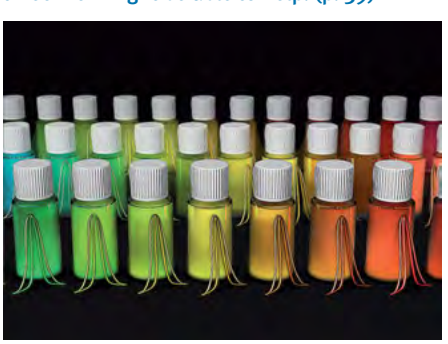


Photo: MIT

Carbon quantum dots are the latest species to emerge from the carbon nanomaterials zoo. Their tunable fluorescence makes them interesting candidates for cell labelling. (p. 52)

News

Opinion

Analysis

Journal Club

Publication Statistics

Biobusiness

Service

Careers

Humour

eppendorf

Order your sample at
www.eppendorf.com/5mL

Your Turn

Eppendorf Tubes® 5.0 mL - now with screw cap

Choose the optimal Eppendorf Tubes 5.0 mL for your laboratory needs: Whether centrifugation, incubation, storage, or other applications - the Eppendorf 5.0 mL system provides the ideal solution for handling samples in the low to medium volume range.

> High g-safe® stability enables safe and fast centrifugation steps
> The new screw cap combines user-friendly handling with an exceptionally tight seal
> Highest sample integrity as no slip agents, plasticizers, or biocides are used during manufacturing

www.eppendorf.com/5mL

Eppendorf®, the Eppendorf logo, g-safe® and Eppendorf Tubes® are registered trademarks of Eppendorf AG, Germany.
U.S Design Patents are listed on www.eppendorf.com/ip. All rights reserved, including graphics and images. Copyright © 2016 by Eppendorf AG.

Picture of the issue

Weird Science

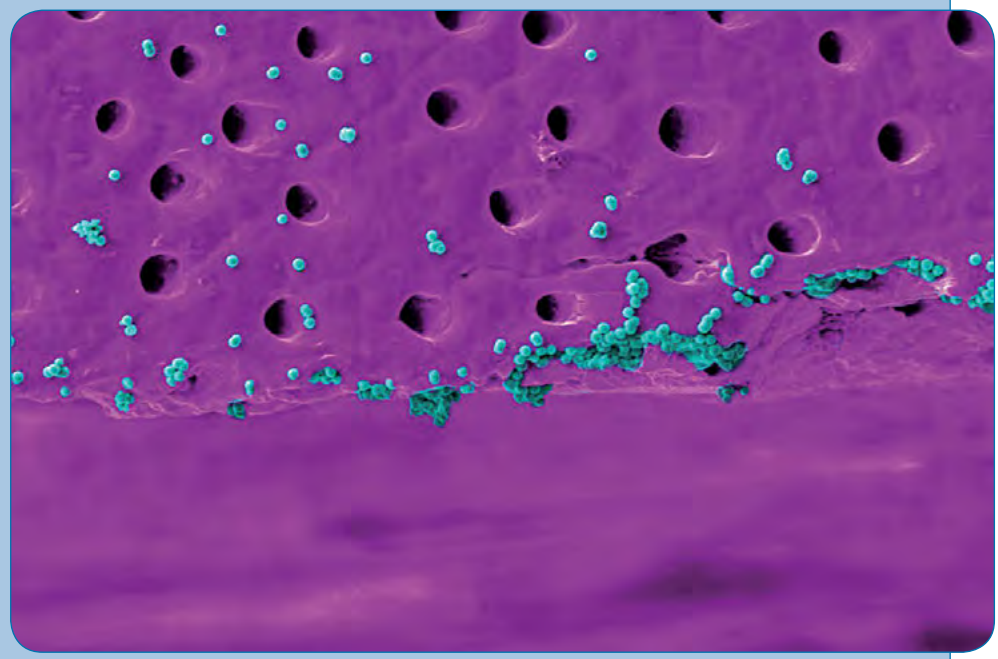


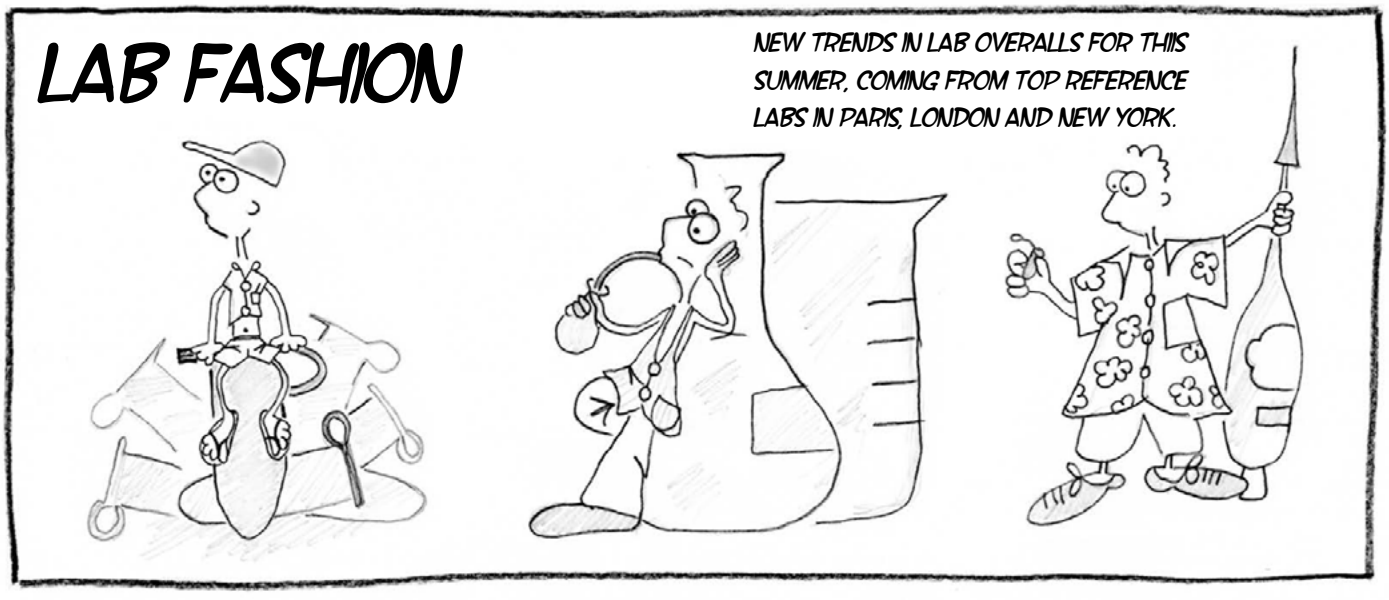
Photo: Nina Vyas, University of Birmingham

It's not often that one is honoured for something weird. And can it get any weirder than nano-dentistry? Not much. That's perhaps why Nina Vyas, doctoral student at the University of Birmingham, won second place in the Weird and Wonderful category of the annual science photo competition, organised by the UK-based Engineering and Physical Sciences Research Council, EPSRC. Her false-colour electron micrograph image (see right), titled On the Edge of Glory, shows parts of her research project: purple-coloured is the surface of a human tooth with its dentinal tubules, the little green beads are not caries bacteria but silica particles of about 800 nm size. What are those silica particles doing on a human tooth, you wonder? "We are investigating how antimicrobial particles can be put into teeth to kill bacteria that invade the tubules during dental decay (...) We are researching a new way to push sub-micron particles further into the channels, using the large forces generated by cavitation bubbles. These are bubbles that implode on themselves and generate high speed microscopic jets and shock waves. We have shown that the sub-micron particles can be delivered into the tubules after just a one second blast of cavitation bubbles," Vyas explains. Weird and wonderful indeed.

-KG-

PAUL THE POSTDOC

BY RAFAEL FLORÉS



TIB®
MOLBIOL

didn't know
that blue eyes
is just rs12913832

LightSNiP Assays

SNP on Demand

More and more human SNPs are analyzed for their potential association with diseases, risk factors and predispositions.
Our LightSNiP assays are pre-established, probe-based tests using a melting curve to detect sequence variations.
These assays are developed on the Roche LightCycler® 480 system, but can be applied also on other instruments that run a melting curve (guaranteed for all LightCycler® systems 1.x, 2.0, 480, Nano and Roche TaqMan® 48. Some LightSNiP assays have been exemplarily tested to work on Illumina ECCO, Qiagen Rotorgene, Bio-Rad CFX96 and other instruments; please inquire).

Convenient to Appiy

LightSNiP assays come premixed with a standardized protocol. Just reconstitute in water, combine with the Roche master reagent, add samples and start your experiment. LightSNiP assays on plates (arrays) available on request.

Simpie to Order

For ordering use the rs number from dbSNP (NCBI/GenBank®)

tib-molbiol.com/oligoshop/SNP

One vial contains primers and probes for 96 rxns each 20 µl.

USA TIB MOLBIOL LLC	**DEUTSCHLAND** TIB MOLBIOL GmbH	**ITALIA** TIB MOLBIOL s.r.l.	**ESPAÑA** TIB MOLBIOL sl
Email: dna@tibmolbiol.com	Email: dna@tib-molbiol.de	Email: dna@tibmolbiol.it	Email: dna@tib-molbiol.es
Tel. +1 (877) 696-5446	Tel. +49 30 78 79 94 55	Tel. +39 010 362 83 88	Tel. +34 91 344 6642
Fax +1 (877) 696-5456	Fax +49 30 78 79 94 99	Fax +39 010 362 19 38	Fax +34 91 344 6670

SimpleProbe® and LightCycler® are trademarks from Roche. SimpleProbe® probes under license of Roche Diagnostics GmbH (for research use only). Homogenous amplification methods with real-time detection are covered by patents owned by Roche Molecular Systems, Inc. and F. Hoffmann-La Roche Ltd. Use of these methods requires a license.

11.006 sign-berlin.de

WWW.TIB-MOLBIOL.COM

Recently Awarded

▶ Not less than five researchers have been honoured with the **Canada Gairdner International Award** for their work on the CRISPR-Cas system. Besides the usual suspects, **Jennifer Doudna** (UC Berkeley), **Emmanuelle Charpentier** (MPI for Infection Biology, Berlin) and **Feng Zhang** (Broad Institute of MIT), who were awarded for the development of the bacterial defense system as a genome editing tool, two more early "Heroes of CRISPR", **Philippe Horvath** (DuPont, France) and French-born, **Rodolphe Barrangou** (North Carolina State University), received the prize for their studies, focussing on the actual immune function of the system in bacteria.

▶ This year's **EMBO Gold Medal** went to two young scientists with extraordinary research subjects. **Richard Benton** (University of Lausanne) was honoured for his work on insect olfactory perception. The scientists is particularly interested in insect odorant receptors. The second prize winner, **Ben Lehner** from the Centre for Genomic Regulation in Barcelona, discovered that phenotypes cannot only be traced back to an individual's genetic make-up and the influence of environmental factors. Phenotypic diversity also arises from "stochastic variances in gene expression early in development." The Medal carries an honorarium of €10,000.

▶ Eppendorf does not only make handy tubes for the molecular biology lab, it also, annually, honours young biomedical scientists with its **Eppendorf Award for Young European Investigators**. This year, the award, which includes a prize money of €20,000, went to **Adrian Liston** from the University of Leuven. The Australian/British scientist studies the adaptive immune system, in particular regulatory T-cells, or Tregs. These cells help the body maintain a healthy balance between autoimmunity and immunosuppression. In his more applied immunological research, he identified several new primary immunodeficiencies and inflammatory diseases. -KG-

New ESFRI Roadmap published

Strategic Plans

In case you missed it, already in March the European Strategy Forum on Research Infrastructures, or ESFRI, published its latest Roadmap (*www.esfri.eu/roadmap-2016*). In it, they announce, which pan-European research infrastructures (such as computing facilities, repositories or libraries) are especially important for the European Research Area and thus, deserve funding (giving a nudge to EU financiers). "The future prosperity of Europe", ESFRI reasons, "in an increasingly competitive, globalised and knowledge-based economy, depends upon fully exploiting the continent's potential for scientific and technological innovation." And this can only be achieved by funnelling powers.

Currently, 21 projects from all walks of science enjoy ESFRI support, which allows them to gradually grow from incubation phase to fully operating "hubs of scientific excellence", benefitting all European scientists. ESFRI follows the infrastructures from its initial, preparatory phase for a maximum of ten years. After which, the projects are expected to "start implementation and reach sustainability for long term operation, thus assuring maximum return on investment in terms of science, innovation, training, socio-economic benefits and competitiveness".

Expect slightly better equipment at the soon-to-be-opened EURO-BioImaging infrastructure.

Photo: www.publicdomainpictures.net/Lynn Greyling

Six new infrastructures entered the new ESFRI Roadmap that "fill in important gaps in the European science landscape". Only one is relevant for life sciences: EMPHASIS (European Multi-Environment Plant Phenotyping and Simulation Infrastructure, which is not be confused with the EU-funded Effective Management of Pests and Harmful Alien Species: Integrated Solutions project or the also EU-funded Explosive Material Production Hidden Agile Search and Intelligence System.

When completed, the *ESFRI-supported* EMPHASIS project wants to back European plant and agricultural science "by developing and providing access to infrastructures addressing multi-scale phenotyping for analysing genotype performance under diverse environmental conditions". Coordinating country is Germany, the Forschungszentrum Jülich to be exact. Costs in the construction phase are estimated to be in the €70 million range. Operation of the infrastructure is scheduled for 2020, with operational costs of €3.6 million per year.

Mere months away from providing its services to European scientists is the EURO-BioImaging infrastructure, which entered the ESFRI Roadmap in 2008. With three imaging facilities scattered throughout Europe (University of Turku, Finland; University of Torino, Italy; EMBL Heidelberg, Germany), the infrastructure wants to give all European scientists the possibility to use state-of-the-art imaging technologies. "Such an open access model will not only bring scientific benefits. It could mitigate the high costs of innovative imaging technologies and the scarcity of expert staff, increase international cooperation and boost transfer of knowledge among European researchers," the coordinators hope. The annual budget for EURO-BioImaging is €1.55 million.

Call for Action on Open Science

Ambitious Goals

The signs bode well for widespread open access to scholarly research. Although it requires not less than a transformation of century-old practices in academic publishing, by 2020, all publicly funded scientific publications should be available to the public as well as the scientific community free of charge. That's one of two important pan-European goals, the Amsterdam Call for Action on Open Science wants to attain. The other one is a "fundamentally new approach towards optimal reuse of research data".

In April, experts and stakeholders came together to discuss the future of academic publishing at the Open Science – From Vision to Action conference. The result is the Call for Action, a "living document reflecting the present state of open science evolution". In it, the authors identify current problems (for instance, access barriers such as paywalls, embargos and the long time between submission of a manuscript and its publication) and suggest possible solutions (researchers are encouraged to try out new and faster ways of publishing, such as flipped publishing, and should no longer accept disclosure clauses). ▶▶

EMBL 2016

Conferences

24 - 26 JUL | EMBL Conference
Microfluidics 2016
C. Merten, S. Quake
EMBL Heidelberg, Germany

27 - 30 AUG | EMBL Conference
Transcription and Chromatin
D. Duboule, E. Furlong, A. Shilatifard, M. Timmers | EMBL Heidelberg, Germany

31 AUG - 3 SEP | EMBO Conference
Chemical Biology 2016
M. Köhn, J. Overington, C. Schultz
EMBL Heidelberg, Germany

7 - 10 SEP | EMBO | EMBL Symposium
Actin in Action: From Molecules to Cellular Functions
B. Baum, J. Faix, P. Lenart, D. Mullins, F. Nedelec, C. Sykes | EMBL Heidelberg, Germany

14 - 17 SEP | EMBL–Wellcome Genome Campus Conference
Proteomics in Cell Biology and Disease Mechanisms
A.-C. Gavin, A. Lamond, M. Mann | EMBL Heidelberg, Germany

25 - 27 SEP | EMBL–Wellcome Genome Campus Conference
Big Data in Biology and Health
E. Birney, B. Grossman, J. Korbel, C. Relton
EMBL Heidelberg, Germany

5 - 8 OCT | EMBO | EMBL Symposium
The Complex Life of mRNA
A. Ephrussi, N. Sonenberg, J. Steitz, D. Tollervey | EMBL Heidelberg, Germany

12 - 15 OCT | EMBO | EMBL Symposium
Organoids: Modelling Organ Development and Disease in 3D Culture
M. Bissell, J. Knoblich, E. Schnapp
EMBL Heidelberg, Germany

19 - 23 OCT | EMBO Conference
Experimental Approaches to Evolution and Ecology Using Yeast and Other Model Systems
J. Berman, M. Dunham, J. Leu, L. Steinmetz
EMBL Heidelberg, Germany

12 - 15 NOV | EMBO Conference
From Functional Genomics to Systems Biology
E. Furlong, F.C.P. Holstege, N. Rajewsky, M. Walhout | EMBL Heidelberg, Germany

20 - 23 NOV | EMBO Conference
Molecular Machines: Integrative Structural and Molecular Biology
J. Briggs, T. Carlomagno, G. Kleywegt, D. Panne, D. Svergun | EMBL Heidelberg, Germany

4 - 6 DEC | EMBL–Wellcome Genome Campus Conference
Target Validation Using Genomics and Informatics
E. Birney, C. Fox, M. Fergusson, S. John
EMBL Heidelberg, Germany

Courses

28 AUG - 5 SEP | EMBO Practical Course
Cryo-Electron Microscopy and 3D Image Processing
J. Briggs, B. Boettcher, L. Passmore, C. Sachse, H. Stahlberg | EMBL Heidelberg, Germany

29 AUG - 2 SEP | EMBL Course
Chromatin Signatures during Differentiation
J. Dreyer-Lamm, P. Grandi, K.-M. Noh
EMBL Heidelberg, Germany

12 - 14 SEP | EMBL-EBI Course
Metagenomics Bioinformatics
H. Denise, L. Emery, A. Mitchell
EMBL-EBI Hinxton, UK

12 - 20 SEP | EMBO Practical Course
Protein Expression, Purification and Characterisation
C. Loew, R. Meijers, A. Parret | EMBL Hamburg, Germany

19 - 23 SEP | EMBL-EBI Course
Structural Bioinformatics
T. Hancocks, G. Kleywegt, C. Orengo
EMBL-EBI Hinxton, UK

19 - 24 SEP | EMBL Course
Extracellular Vesicles: from Biology to Biomedical Applications
J. Dreyer-Lamm, A. Hendrix, E. Nolte-'t Hoen
EMBL Heidelberg, Germany

3 - 6 OCT | EMBL-EBI Course
Introduction to Next Generation Sequencing
T. Hancocks, J. Randall, M. Rossello
EMBL-EBI Hinxton, UK

4 - 7 OCT | EMBL Course
Whole Transcriptome Data Analysis
V. Benes, R. Calogero | EMBL Heidelberg, Germany

16 - 23 OCT | EMBO Practical Course
High-Throughput Microscopy for Systems Biology
J. Ellenberg, D.W. Gerlich, B. Neumann, R. Pepperkok | EMBL Heidelberg, Germany

17 - 24 OCT | EMBO Practical Course
Solution Scattering from Biological Macromolecules
A. Kikhney, D. Svergun
EMBL Hamburg, Germany

7 - 11 NOV | EMBL-EBI-Wellcome Genome Campus Course
Resources for Computational Drug Discovery
T. Hancocks | EMBL-EBI Hinxton, UK

9 - 10 NOV | EMBL Course
Microinjection into Adherent Cells
R. Pepperkok, S. Terjung, S. Stobrawa
EMBL Heidelberg, Germany

21 - 25 NOV | EMBL Course
NGS: Enrichment Based Targeted Resequencing
V. Benes, J. Dreyer-Lamm, A. Heim
EMBL Heidelberg, Germany

28 - 29 NOV | EMBL Course
NGS: Whole Genome Sequencing Library Preparation
V. Benes, J. Dreyer-Lamm, A. Heim
EMBL Heidelberg, Germany

28 NOV - 2 DEC | EMBL-EBI Course
Biological Interpretation of Next Generation Sequencing
G. Rustici | EMBL-EBI, Hinxton UK

4 - 9 DEC | EMBL-EBI-Wellcome Genome Campus Course
Proteomics Bioinformatics
L. Emery | EMBL-EBI, Hinxton UK

7 - 11 DEC | EMBL Course
Microbial Communities: Modelling Meets Experiments
R. Mahadevan, K. Patil, K. Sasaki
EMBL Heidelberg, Germany

For full event listing please visit our website
www.embl.org/events
@emblevents

We would like to thank the members of the EMBL ATC Corporate Partnership Programme:
Founder Partners: Leica Microsystems, Olympus
Corporate Partners: BD, Boehringer Ingelheim, GSK, Illumina, Thermo Fisher Scientific
Associate Partners: Eppendorf, Merck, Nikon, Sanofi

EMBL

"To achieve these objectives, all of the stakeholders must take action. Scientists' evaluation and assessment systems will need to be reviewed. Universities and research funders must ensure that the new system values the impact of research and sharing results more highly, instead of just looking at numbers of publications and citations. The EU member states and the European Commission must better align their policies to facilitate open access and open data, thus making policy more uniform," the Call urges.

Several European nations have already done their homework. Switzerland is currently preparing its national Open Access strategy, as suggested by the Amsterdam Call. And the country's principle public funding body, the Swiss National Science Foundation (SNSF), recently released a monitoring report, revealing that "40 per cent of publications produced with the aid of SNSF funding are openly accessible". Proudly, the SNSF announces that this "impressive figure is set to rise" even further.

In related Open Access news, the Directory of Open Access Journals (DOAJ) recently purged its inventory. The DOAJ operators removed more than 3,000 journals that did not renew their application, even after being reminded three times. "We are confident that the majority of the journals removed have never supplied article metadata to us, or have done once but haven't sent us anything for at least two years." Currently, the database gives access to more than 8,000 Open Access journals and close to two million articles.

Research vessel naming contest

Ship Ahoy!

More famous than Lady Gaga and Angela Merkel together, Boaty McBoatface took the world by storm. With overwhelming majority, the entry submitted by *BBC* radio presenter James Hand won the Name Our Ship competition by the Natural Environment Research Council (NERC). Was it all a clever publicity stunt by the NERC or just a scientific outreach activity that has gotten totally out of control?

Fact is, the £200 million research vessel will, after all, cruise Arctic and Antarctic waters not as Boaty McBoatface but under the name "RSS David Attenborough". Appreciating the public's "truly inspirational and creative" name suggestions, UK science minister, Jo Johnson, ultimately vetoed against Boaty. For Attenborough, it's, of course, an honour. "I have been privileged to explore the world's deepest oceans alongside amazing teams of researchers, and with this new polar research ship they will be able to go further and discover more than ever before," he told the *Guardian*.

At the beginning of May, the men behind the Name Our Ship competition also had to report to the House of Commons as part of a general inquiry into science communication in the UK. Duncan Wingham, NERC's head, confessed that although the contest had run out of the rudder, "I would like to think [the minister] sees this as what I'd describe as an incredible success (…) We could make the claim that we're probably now the best known research council in the world. Hundreds of thousands of people now know not only about us, but about the science we have done."

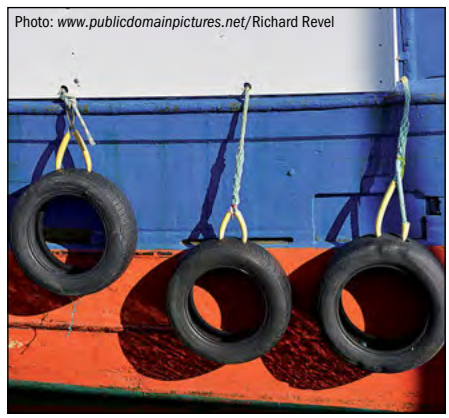


Photo: *www.publicdomainpictures.net*/Richard Revel

Indeed, with their contest, the Council reached not less than 214 million people on Twitter alone. Thousands watched the Council's videos on Youtube and read about their scientific projects. A dream come true for any marketing strategist.

The new vessel, the UK's "largest and most advanced research ship yet", will set sail to the Antarctic in 2019. Carrying nine double-decker busses worth of scientific equipment, the RSS David Attenborough will help researchers find out more about, amongst others, the stability of the Antarctic ice sheet and the diversity of marine life.

And what about Boaty McBoatface? According to the NERC, the name "will live on as the name of the ship's high-tech remotely operated sub-sea vehicle". Reporting to the Members of Parliament, Duncan Wingham said that the Council did not yet have time to come up with an appropriate media strategy for the bright yellow vehicle. The public can, however, be sure that it will re-surface very soon.

EU regulation of EDCs

Basic Agreement

It's a hotly fought over topic – are endocrine disrupting chemicals (EDCs) harmful for our health and need strict regulation? Or can they, at low concentrations, safely be tolerated? For years, the conflict between different pressure groups has been simmering in Europe. In April, 23 scientists, moderated by former European Science Advisor, Anne Glover, and former European Commission President, Jose Manuel Barroso, met in Berlin to, for starters, come to a mutual agreement about clear "criteria for identifying the hazard potential of harmful endocrine substances". These criteria are just the very first but important step in the entire risk assessment procedure.

Indeed, the participants arrived at a joint solution. In the resulting Consensus Document, the scientists, including Andreas Kortenkamp (Brunel University London), Åke Bergman (Swedish Toxicology Sciences Research Center) and Alberto Mantovani, (Istituto Superiore die Sanitá, Rome) recognise that "the identification of chemicals that contribute to adverse effects on human health is fraught with difficulties, which, in the case of endocrine disruptors, can be traced to several specific factors". For instance, exposure to EDCs often occurs during puberty and thus, effects are "difficult to re-construct", the scientists write. In addition, "internationally validated test systems" are yet to be optimised. "We believe that a consensus about these issues is unlikely to emerge in the near future. Nevertheless, in our view, the establishment of criteria for the identification of endocrine disruptors is possible without resolution of these issues."

Andreas Hensel, president of the German Federal Institute for Risk Assessment, which organised the meeting, called the Consensus Document a "breakthrough". "The results can support the European Commission in taking science-based measures where required to reduce endocrine disruptors, for instance in consumer products, pesticides and also in food."

Despite this laudable basic agreement, the opposing camps continue on their path. Seven Concerned Toxicologists for Better Science and Regulation recently approached the EC Commissioner of Health & Food Safety, Vytenis Andriukaitis, to lobby for their cause. During a meeting in Brussels, Colin Berry, Alan Boobis, Wolfgang Dekant, Daniel Dietrich, Helmut Greim, Pat Heslop-Harrison and Richard Sharpe – ▶▶

FARMACUBA

Empresa Importadora-Exportadora
Farmacuba, integrada a la OSDE
BioCubaFarma.

FARMACUBA, SALUD AL ALCANCE DE TODOS

Importa materias primas, envases, reactivos, equipos y piezas de repuesto para la industria farmacéutica cubana y medicamentos para el Sistema Nacional de Salud de Cuba.
Exporta medicamentos genéricos y servicios tecnológicos para la producción de medicamentos.

Calle 70 Nro. 29B14 entre 29B y 29C. Playa. La Habana. Cuba.
Teléfonos: (53) 7212 5796 al 99 Pizarra
Dirección: Telefax: (53) 7204 8278, E-mail: direccion@farmacuba.cu
Importaciones: Telefax: (53) 7204 7257, E-mail: importaciones@farmacuba.cu
Exportaciones: Telefax: (53) 7204 7258, E-mail: exportaciones@farmacuba.cu

BioCuba Farma
GRUPO

Boobis, Dietrich and Greim had signed the Consensus Document – told Andriukaitis of their fear that public opinion about EDCs is currently "dominated by certain scientists, NGOs and well-funded pressure groups, who categorically assert that EDCs contribute to human cancer, reproductive disorders, obesity and type 2 diabetes". There's an "onslaught of pseudoscience in the EU that needs to be stemmed", they claim.

New visa directive for non-EU scientists

Attractive Effect

Some build fences; others welcome multicultural influences from around the world. In May, the European Parliament passed a new visa Directive for third-country nationals, who want to come to Europe for the purpose of research, studies, training, volunteering, pupil exchange and au pairing. The new Directive, which merges two existing directives, has been in the making since 2013, with advocating contributions from, for instance, the European Students' Union and the Erasmus Student Network.

Four points have been negotiated to make Europe more attractive for non-EU researchers and students: firstly, students and researchers may stay at least nine months after finishing their studies or research in order to look for a job or to set up a business; secondly, students and researchers may move more easily within the EU during their stay, they will not need to file a new visa application but only notify the member state, to which they are moving and researchers will be able to move for longer periods than currently allowed; thirdly, researchers have the right to bring their family members with them and these family members are entitled to work during their stay in Europe, and, last but not least, students have the right to work at least 15 hours a week.

"I am glad that the EU recognises the value of attracting highly skilled people to come here and to entice them to stay by creating a harmonised European system applicable in all member states," said lead MEP, Cecilia Wikström.

The European Students' Union is also happy about the new Directive but, for them, it's not going far enough. "The final document represents half of what we would have loved to see, as the proposals from the European Parliament were way more advanced in rights for non-EU students, but still, some of our postulates have been watered down," says Fernando Galan, the European Students' Union's Chairperson, in a press release. For example, the Parliament had proposed an 18-month-stay for researchers after finishing their studies and a maximum of 20 hours of work per week for students. Therefore, the ESU hopes that this "Directive is only a first step in ensuring that Europe attracts more non-EU students to its campuses and encourage and increase internationalisation".

After the Directive enters into force, member states have two years to implement it.

-KG-

Move to the Beat

UK physicists find that unicellular algae's swimming style depends on intracellular structures.

It's already astonishing that simple, microscopic life-forms like *Paramecium* or *Chlamydomonas* are able to move towards light or food. It's even more astonishing that they do this in a coordinated fashion, perfectly synchronising their hair-like, cellular appendages, the flagella or cilia. For many decades, scientists wondered how this synchronisation, resulting in directed locomotion, instead of pointless staggering like a drunk at closing time, is achieved. Hydrodynamic interactions has been their answer – fluid flows passively drive the coupling of flagella and cilia.

This might be true for some algae like *Volvox* but it can't explain all gait types of unicellular algae. The green alga, *Chlamydomonas reinhardtii*, for instance, synchronises its two flagella in such a way that it moves with a perfectly executed breaststroke swimming style. Hydrodynamic forces alone can't be behind this type of locomotion. There must be something inside the cell, physicists Kirsty Wan and Ray Goldstein at Cambridge University speculated. "Pairs of interacting flagella evoke no image more potent than Huygens' clocks: Two oscillating pendula may tend toward synchrony (or antisynchrony) if attached to a common support, whose flexibility provides the necessary coupling," the scientists write in their latest paper (*PNAS*, 113(20):E2784-93).

Indeed, already back in the 1960s, scientists noticed contractile fibres, connecting pairs of basal bodies in the flagellar apparatus. Basal bodies anchor the flagella and function as centrioles during cell division. Among the fibres connecting *Chlamydomonas reinhardtii*'s basal bodies is the bilaterally symmetric distal fibre (DF) that attaches to the basal bodies' middle section and lies exactly in the plane of flagellar beating. A second set of fibres (parallel proximal fibres, PF) holds together the BBs at one, the proximal, end. "The distal fibre likely couples *Chlamydomonas reinhardtii* flagella, providing a degree of freedom that can reorient a flagellum at the BB through its contraction," Wan and Goldstein suggest. Interestingly, other unicellular algal species with different gaits, such as galloping and trotting, possess a differing basal architecture (BBs and connecting fibres) that matches their type of locomotion.

Experiments with mutants and micromanipulation are in favour of the physicists' hypothesis. Observing the motility of the vfl3 mutant of *Chlamydomonas* with missing, misaligned or incomplete distal fibres, they saw aimlessly floundering algae, "rotating in place at the chamber bottom". Likewise, when the scientists trapped one of *Tetraselmis suecica*'s flagellum, a marine green alga, the remaining three flagella simply continued their coordinated beating pattern, resulting in a transverse gallop gait, as though nothing had happened.

"Until now there have been many competing theories regarding flagellar synchronisation, but I think we are finally making sense of how these different organisms make best use of what they have," commented Goldstein in a press release. -KG-

(More research results from European labs on pp. 28-33)

Photo: Ruhr Universität Bochum, AG Photobiotechnologie

Observations of the Owl (60)

Bossy Bosses

Okay, I admit it: we owls tend to be rather bossy birds. Is this not a natural consequence of being as brainy as we owls undoubtedly are? Full of *wisdom*, as even you humans never tire of declaring.

Anyway, it reminds me of a certain episode, when I was a young professor down in the southern mountain woods. Yes, at *that* time, I was a real showpiece of a "bossy boss". I viewed it to be a downright given law that *I* was the one to dictate to the young birds in *my* lab exactly what ideas to follow and even what precise experimental approaches to take. I just couldn't imagine that one of those wet-feathered greenhorns might come up with any valuable ideas of their own. And indeed, none of the awestruck birdies ever even dared to question my authority. Until one particular day in the cool-cave, deep underground…

I cannot remember exactly why I entered the cool-cave on that day. Anyway, I had just fluffed up my feathers to cope with the underground chill, when I realised that in the opposite corner of the cave, young Crow was busily transferring quite a number of samples with his beak, wings and claws into a complicated machine. For quite a while, I just kept silently watching him and tried to guess what on earth he was actually doing there.

"None of the awestruck birdies ever even dared to question my authority."

We had been talking about binding assays, I recalled. (Of course, *I* had actually *told* him to perform a certain series of binding assays.) But what he was doing right here, right now was definitely *not* a binding assay, I realised, perplexed.

A couple of minutes later, Crow, with a deep breath, finally pushed the start button. I waited for another short while before I moved. Immediately Crow turned around, startled, "Oh, er, hello, Owl. What are you doing down here in these dark depths?"

I forget my exact answer to this question but I remember asking in the most innocent-sounding voice that an owl could ever muster, "Any news on your binding assays, per chance?"

Crow rolled his eyes, "Yes, I am almost done with the assays you suggested. But no effects, so far."

"No effects," I stupidly repeated. "Hmm, I shall give that some thought when back up on the warmth of my branch. But tell me… this machine you've just loaded – it definitely can't perform binding assays, can it?"

"No," was Crow's short reply.

I waited for a further explanation but Crow just kept silently staring at me with his dark, glittering eyes.

"Hrrm," I finally uttered. "Perhaps you would be so kind as to explain to me what you are actually doing."

"Trying a different approach," he cryptically replied.

"A different approach. Well, well…" I ruffled my feathers agitatedly. "C'mon, Crow. A bit more please! Or do you think I don't have to know *any*thing about what *my* students do in *my* lab with *my* grant money?"

"I am trying to purify the binding protein," Crow replied. His voice still remained provocatively calm but his dark eyes seemed to send out small bright flashes.

"Purify the binding protein?" Apparently, I seemed to be having a "polly-parrot" day. "Hadn't we both agreed that we'd learn the most about the signalling mechanism by studying the binding activity of the extract under a whole variety of different conditions? After all, it's mechanisms that count – not single proteins!"

"Right." Crow loosened his stare. "And I have been doing tons of binding assays lately, but haven't got a single step further. Believe me, Owl, we won't get ahead with this approach! 'Try the same but harder' has never been a promising strategy in science. If you get stuck you'll *have* to try something *new*."

"Purify the binding protein." I was definitely stuck in stupid-repetition-modus.

"Yes, Owl. It's not without reason that protein experts say 'structure follows function'. Please, let me purify the binding protein and study its structure – I'm sure that we'll get valuable clues as to what kind of signalling mechanism it triggers."

"And that's what you are doing here: purifying the binding protein?" Wow, repetition of the repetition!

"Exactly. I know, I should have told you earlier but be honest, Owl, you would have shot down my suggestion right from the start. So, I thought I could perhaps convince you better with some promising first results at hand. Actually, I still can't show you much right now but I have a strong feeling that *this* approach will indeed work. C'mon, Owl, please let me continue."

Never before had a student spoken to me like this before – and thereby clearly expressed that *my* approach was chicken-shit. Something must have gone "klick" within and I almost couldn't believe my own ears, as I agreed. "Okay, sounds reasonable. Go ahead with it. But please keep me up to date."

Today, I know that *this* was the key moment for me to understand that, in contrast to my former "bossy" attitude, it is one of my highest duties as professor and supervisor not only to *allow* but even to actively *encourage* my students to follow their own ideas and notions. Of course, they might fail here and there – but in the end, it makes them better scientists.

Crow, by the way, finally unravelled the whole signalling mechanism by following his instinctive approach – and proved himself to be one of the best students I ever had.

Three weeks ago, I happened to meet *Professor* Crow at the Western Lowland Congress Grove where he told me how his new lab is running. And I was greatly astonished to realise that, in the meantime, he appears to have evolved into quite the "bossy boss" professor himself – despite his own former experiences in my lab…

Comments: owl@labtimes.org

Research Letter from:… Greece

Therapeutic Dance Jumps

By our corresponding author, **Efthimos Altis**

Financial crises in Greece have resulted in diminished research funds. In such circumstances, it is perhaps not surprising that some medical researchers have sought therapeutic solutions that do not require expensive technology and overpriced medications. For example, Zacharias Vordos who has looked at the "Impact of traditional Greek dancing on jumping ability, muscular strength and lower limb endurance in cardiac rehabilitation programmes" (*Eur J Cardiovasc Nurs*, 2016 April 14, Epub ahead of print). Vordos says that Greek dancing is an important part of weddings and other celebrations, it represents the culture of the country and is a popular, inexpensive activity among both young and old people. He also claims to show that it can improve the health of elderly cardiac patients. "Patients who participated in Greek dancing jumped higher, probably because they had stronger leg muscles."

Based at Aristotle University's Laboratory of Sports Medicine in Thessaloniki, Vordos' study aimed at an alternative form of therapeutic exercise for forty Greek patients recovering from chronic heart failure (aged 73.2±4.7 years). "The jumping ability in older people with heart failure has not previously been evaluated," he notes. In fact, none of his patients had participated in any form of exercise during the previous year, let alone jumping.

They were assigned to a dance Group A (13 men and seven women) or to a control Group B (14 men and six women). Patients in Group A followed a 12-week exercise training programme consisting of three 40–65 min sessions per week: after a 10 minute warm-up, they performed Greek traditional dances. Meanwhile patients in Group B were asked to continue their "sedentary lifestyle".

Photo: Ivorygirl, CC-BY-SA-3.0,2.5,2.0,1.0

Get jumping

The exercise capacity of the patients was evaluated before and after the 12-week dance programme: How far could they walk in six minutes? How strong were their leg muscles? And, most importantly, how high could they jump in "plyometric, countermovement, and squat jumps"?

After their dance therapy, Vordos says Group A improved "significantly" compared to Group B. They walked further – an extra 48 metres (+9.6%) – and jumped higher – an extra 1.5 cm (+12.7%), while Group B showed no change.

Vordos says the patients were randomly assigned to the two groups. However, the data in Table 2 clearly shows that his dancing Group A were fitter and stronger than Group B. At the start of the experiment, patients in Group B walked 36 metres less in six minutes (-7.2%), had weaker legs (-10%), and jumped lower in all three jumps (-9.2%).

Furthermore, Vordos says all the patients were "pre-informed" about the purpose and procedures of the study, that is, they knew it was designed to show that Greek dancing was good for their recovery. A possible source of jumping bias?

It might also be noted that the only control is total inactivity. One could argue that any non-strenuous physical activity would be of benefit but that only Greek dance was tested. It was not compared to another exercise regime, e.g. a 30 minute walk in the park each day. This does not sound like such a randomly "controlled trial".

Nevertheless, Vordos happily declares that "Greek dances have a positive effect on jumping ability in patients with chronic heart failure". He concludes ,"Traditional Greek dancing is enjoyable and sociable, and we have now shown that it leads to health benefits in elderly patients."

A world ranking in national folk dance?

But Vordos does concede that other dances may also provide "cardiac benefits", e.g. "Zumba fitness programmes with Latin music". This also provides possibilities for further research. Just think of all the other national folk dances we could test therapeutically – a medical folk dance ranking!

Already in Italy, researchers have tested the benefits of "waltz dancing" for cardiac patients (*Circ Heart Fail*, 1(2):107-14). While in Turkey, they found that "Turkish folklore dance" improves "balance, depression and quality of life in older women" (*Arch Gerontol Geriatr*, 48(1):84-8). Meanwhile, in the US, they compared the benefits of "Tango dance" for patients with Parkinson's Disease (*Front Aging Neurosci*, 7:239). The islanders of Hawaii have reported on the cardiac benefits of dancing "Hula" (*Health Promot Pract*, 16(1):109-14). And traditional Thai dance apparently has "cardiopulmonary effects" on menopausal women (*J Phys Ther Sci*, 27(8):2569-72).

Meanwhile, in the Netherlands, possibly for lack of traditional dance steps, they have investigated the possibility that people with osteoporosis (fragile bones) might be less likely to break them if only they could learn to fall over more gracefully – martial arts style (*BMC Res Notes*, 3:111). But teaching kung-fu rolls to old ladies with brittle bones has its drawbacks. This Dutch study wisely concluded that it might be better if people train to fall when they are still young and healthy, i.e. many years before osteoporosis sets in.

Therefore, future dance research might also consider healthy people whose prospective disease condition has not yet developed. Who knows, perhaps stressed and undervalued researchers might also benefit from a bit of relaxing social exercise. Care for a dance…?

What's une Femme to Do?

Nero,

The summer season has begun the annual rotation of nation-defining events. The right royal HR Lizzies's royal approved: Royal Flower Show, Royal Ascot Horse Racing and Royal Henley Regatta. These are the must attend events frequented by the tweed wearing, brogue stomping establishment, in which they remind us how the other half lives. Alas, disharmony has broken out as the two leading Old Etonians Tweedledee (David "the Camera's On") and Tweedledumb (Boorish Johnston) are at logger heads. The man who is and the man who would be King are arguing over chips and pommes frites. These once single club buddies are the self-proclaimed lead agitators for the arguments to encourage the Island nation to retain or quit their formal association with the Common Market. Should Britain stay or Brexit? The Polls; the pre-election estimates rather than a mis-spelling of the noun used to describe our carpenters and plumbers, suggest it will be a tight call. It is far from clear if Dave or Boorish will win the day. It really seems that we can't decide.

Neck and neck

They've tried everything suggesting, we'd be millions of pounds or euros better off. They have differently argued we will have cheaper or dearer houses but whether or which of these would be good is unclear. They have oppositely argued that a vote in their direction makes paralysing cyber or Martian attacks more or less likely. It even seems that the outcome will define if Ingerland will ever win another soccer ball world cup or not (*The Guardian* Online, Sept 11th, 2015). However, these various mixed messages seemed not to be tipping the balance. Rather, the fear of the growing influx and influence of Birkenstocks and various sandal wearing behaviours will define the outcome of this historic vote on June 23rd.

The empire and the British way of life was, of course, built upon the sock and enclosed shoe sandal. These would be worn with the full baggy shorts. This is a look cultivated by Lord Baden-Powell, the founder of the Boy Scouts and veteran of Mafeking, who developed this as the uniform of the Brit abroad. An ankle sock would be okay but the look that best advertised British independence required a full knee-high sock. This military-inspired summerwear ensured that the British could advertise their superiority over the Hun, Wops or any other European cousin. Yes, despite humidity and the chaff- and thrush-inducing sweat that the socked sandal combo propagated, this is how Ingerlish cruised around the European tourist hotspots. A growing number of young compatriots, however, noted that there might be some sense in the European way. We had our eyes opened to sandal El Fresco, in which toes and foot were naked. In half a generation, we had gone full continental and allowed the naked sandaled foot to immigrate into our Sceptre Isle.

The proponents of the Brexit are concerned by diminution of Ingerlish culture. However, I feel they have a particular antipathy to the Teutonic podiatry architecture that is the Birkenstock. Now, this rubber-cork-leather bound sandwich moulds the foot ergonomically. It imparts both ventilation and foot-based agility. A critique of this hulking foot timber, whether in its single, double or ankle-strapped variety, was that it lacked the daintiness of high fashion. But time has established it as a classic for young and old. This is foot infrastructure of the highest order and led a revolution in infiltration into British culture. The health and efficiency of the open-toe sandal has been embraced by many Brits and undermines the sensibility of the proponents of Brexit.

Wellingtons for Waterloo

The foreign, open-toed culture could not be tolerated, so the Brexit proponents have evoked the spirit and the ghost of Wellington. Not the General, who defeated Napoleon and halted a previous European invasion at Waterloo. No, Wellington of the rubberised boot. Although conventionally used to allow rural outdoor pursuits in the precipitous summers on our Island Nation, the black Wellington boot has more recently been beautified with flying pigs, sunflowers and insundry, thus making it the funkified preferred alternative summer footwear. Young people everywhere will head to the summer festivals wearing the Albion protectors of days of yore. This will promote British spirit and counter the Europhile Birkenstock-wearing, continental wannabees. First, we take the footwear of the young and their minds will follow. Boorish and other True Grit Brexiteers will lead us over the horizon into a dawn of an Uncommon Market. We will become the Nation that used to have the sunsetless empire saying good night to the EU. The advantages remain unclear but our European cousins can look forward to the re-emergence of the *Candida albicans* riddled knee-length, sock- wearing tourist coming to a major European City soon.

Cry, Boris (the Turkish immigrant) for England and Saint George – but not the Portuguese one.

Nero, could you send a job lot of Black Arizona's just in case they run short in the shops.

FintaN

European marine microbiology

Diving for Molecular Treasures

About one million microorganisms are bustling around in one millilitre of seawater. The EU-funded MaCuMBA project aims to uncover their biotechnological and pharmaceutical potential.

Photo: www.publicdomainpictures.net/ Petr Kratochvil

Until now, despite their abundance, tiny marine creatures have been an almost untapped biotechnological resource. Scientists obtained the DNA of some of these organisms, which afforded clear proof of their existence. Further analyses have been hampered, however, by a lack of appropriate methods of isolation and cultivation. The MaCuMBA project, short for '**Ma**rine Microorganisms: **Cu**ltivation **M**ethods for Improving their **B**iotechnological **A**pplications', is dedicated to sampling and cultivating bacteria, fungi, algae, plankton, and viruses from a wide range of marine habitats. The associated researchers and companies from eleven European countries want to uncover their wealth of bioactive metabolites using high throughput techniques. Between 2012 and 2016, the consortium that includes eleven academic institutions, has received financial support from the European Commission of nearly €9m.

Marine microorganisms on chips

The Dutch firm MicroDish BV, one of nine biotech and biopharma companies involved in the project, created new culture chips especially adapted for marine culture and screened marine microorganisms for useful enzymes. Its researchers were particularly successful in growing both new and familiar marine fungi. The MicroDish Culture Chips contain from 3,000 to 240,000 culture wells with micrometre dimensions. They can be used for high throughput screening, imaging and sample recovery. They also allow viable counting, the capture of microorganisms by filtration and the isolation of previously uncultured organisms from complex samples. The porous base of the chips allows nutrients from beneath the chip to supply organisms growing on its upper surface. "The nutrients can be from a natural source, such as sediment, allowing a very close to natural growth situation," explained Colin Ingham, the Chief Scientific Officer. "Our chips are already used by researchers for screening, the creation of sample collections and the printing of replicates of microcolonies. Scientists also grow organisms under extreme conditions on our chips, for example, at high temperature or with a minimum of water."

Bioactive molecules from the deep sea

Actinobacteria are gram-positive microorganisms, which live in marine and soil habitats and can be cultured in fermenters. The phylum, mainly the genus *Streptomyces*, comprises well-known producers of bioactive natural compounds, such as antibiotics and anti-tumour agents. With a large genome, they harbour many genes related to secondary metabolism. Therefore, the MaCuMBA scientists expected to discover antibacterial, antifungal, anticancer and immunomodulatory metabolites by newly-developed culture procedures.

"During the MaCuMBA project, we have explored the isolation, culture and screening of deep-sea marine bacteria, mainly actinobacteria. We have isolated some interesting anti-tumour compounds," said Fernando de la Calle, Head of Marine Microbiology at PharmaMar. The Madrid-based pharmaceutical company provides protocols for the isolation, molecular characterisation, culture and screening of thousands of actinobacteria, and identification of bioactive compounds. "Other types of marine bacteria, such as rare and new proteobacteria, are also exciting candidates for the production of new bioactive compounds," de la Calle observed. In the past, PharmaMar successfully purified a cytotoxic agent from sea squirts. This acts by targeting the transcriptional machinery and by impairing DNA repair. The corresponding drug, Yondelis or trabectedin, has been approved by EMEA for the treatment of soft tissue sarcoma and ovarian cancer. It is also marketed in the USA and Japan for the treatment of soft tissue sarcoma.

Lynn Paterson's group at the Institute of Biological Chemistry, Biophysics and Bioengineering at the Heriot Watt University, Edinburgh, UK, found ways to isolate microbes using the force of light. Paterson is associated with the MaCuMBA work package 'Development of hardware and equipment for high throughput isolation, cultivation and screening'. "We have developed optical tweezers and an optofluidic device, which we will be exhibiting at the final MaCuMBA conference in Berlin. Feedback from the attending scientists will be useful, to determine whether there is a market for our laser tweezers, or whether the community will be better served by offering a cell isolation service," the molecular biologist and Lecturer in Physics said. The optofluidic device is still at the research stage.

Pulling...

The acting force behind the optical tweezers is a laser beam that is tightly focussed through a microscopic objective. The optical force pulls a nearby cell into the beam focus and traps it, so that it can be moved in three dimensions. The scientists chose near infrared laser light to minimise damage to the cells, which can occur by heating the cells or via the generation of free radicals. "We've fabricated bespoke microscope slides with reservoirs, channels and collection chambers, in which to collect and culture individual cells. Supported by our project partners, we have also developed software to make the process more user-friendly and semi-automated," Paterson reported. Using laser tweezers, the scientists could isolate a wide range of lab-grown bacteria as well as microalgae and yeast.

...and pushing under the microscope

The novel optofluidic device makes use of the scattering force of light. Even

when not tightly focussed, laser light can exert a force on particles and make them move by pushing them in the direction of beam propagation. "Much like a football can be pushed in a jet of water from a fireman's hose," Paterson said by way of illustration. The researchers also prepared bespoke three-dimensional microfluidic circuitry with integrated waveguides to control the flow of cells in a sample. For this, they used ultrafast laser inscription and selective chemical etching, a technology developed by Ajoy Kar at Heriot Watt University. "This means we can direct light into the microfluidic channel and, using the scattering force, push selected cells out of the stream in order to isolate them. You can call it 'flow cytometry and flow sorting on a chip'," Paterson explained. The small device has dimensions of 2x3x1 mm. The researchers were able to manipulate microalgae two micrometres in diameter and larger cells, such as mammalian cells. The device still has to be optimised for the sorting of bacteria.

Moreover, scientists associated with the Université de Bretagne Occidentale developed the 'Cocagne platform'. "With this high-throughput system, we can simultaneously culture hundreds of aero-tolerant, non-pathogenic microorganisms in different media and at different temperatures, isolate and identify strains, and screen for substrate degrading or antimicrobial activities," said Gwenaelle Le Blay, leader of the work package 'Improving culture efficiency of already isolated and cultured microorganisms'.

Over 2,000 new strains

In total, scientists associated with the MaCuMBA project have isolated thousands of new strains by various methods. More than 2,000 key strains have been deposited in four main collections: Roscoff Culture Collection (RCC), Leibniz Institute DSMZ – German Collection of Microorganisms and Cell Cultures, Culture Collection Yerseke and Université de Bretagne Occidentale Culture Collection.

The stored microorganisms include eukaryota, bacteria and archaea from coastal waters to the open sea. Even microorganisms from deep-sea sediments and hydrothermal vents have been collected and preserved. "All of these repositories have a section on their website for searching and ordering strains isolated during the MaCuMBA activities. Currently, some of the strains are available only to project members. At the end of the project, they should be made openly available," said Daniel Vaulot, leader of the MaCuMBA work package 'Secure novel bioresources and provide access to genetic and phenotypic information' and Director of the RCC. "Cultures of marine microorganisms are free for MaCuMBA partners. Other customers have to pay a nominal fee. In the case of the RCC, the fee is different for public research organisations and private companies," he added.

Free service

Furthermore, the German company Ribocon, which focuses on computer-based DNA sequence analyses for environmental, clinical and molecular microbiology, developed the JSpeciesWS Online Service for advanced species differentiation. The service is free and results can be used for research, without any restrictions.

"We have improved the growth of several hundred marine microorganisms. Now, we also have proof of the presence of so far unknown microorganisms in a variety of marine environments through metagenomics analyses, including transcriptomics, proteomics and metabolomics approaches. However, the duration of the project is too short to follow all of these leads," noted the project co-ordinator Lucas Stal, Royal Netherlands Institute for Sea Research. "The MaCuMBA consortium has abandoned the old paradigm that microorganisms can only be studied in pure single strain cultures. In nature, organisms do not live alone but in close association with other species, with which they cooperate or compete by communicating and exchanging growth factors," Stal observed. For, example, MaCuMBA researchers at the University of Warwick have discovered that the marine cyanobacterium *Synechococcus* sp. had to be co-cultivated with the marine bacterium *Rugeria* sp. to keep cells of both genera viable in culture for up to six months.

Better growth in co-cultures

The work package 'Cell to cell communication in community cultures, isolation and cultivation' focussed on the diverse microorganisms on marine sponges and their signalling molecules and pathways. "We have uncovered an array of communication signals and their quenching counterparts," reported Fergal O'Gara, work package leader and Director of the Biomerit Research Centre at the University College Cork, Ireland. "These compounds could be used to supplement media in order to enhance the diversity of culturable organisms isolated from the marine environment. The growth of so far unculturable species could be supported by the silencing of growth-limiting signal molecules," the Emeritus Professor of Microbiology suggested. Indeed, transwell co-culture of sponge samples with signal producing or quenching microbial isolates led to more diverse microbial cultures.

In marine sponge microbiota, the scientists discovered signalling molecules, such as alkyl quinolones and identified members of the well-characterised acyl homoserine lactone class of signals. These molecules are used by gram-negative bacteria, which interact with plant or animal hosts, to communicate and regulate collective behaviour dependent on population density, a process called 'quorum sensing'. The molecules induce changes in the expression of genes, which control, for example, bioluminescence, biofilm formation or virulence. Extracts from the novel signal-quenching isolates interfered with biofilm formation, a key marker for microbial pathogenesis and persistence. "Small molecule mimics from marine organisms that block the formation of biofilms could re-establish the effectiveness of conventional antibiotics," O'Gara said.

At their annual general assemblies in Roscoff, France, Cadiz, Spain and Reykjavík, Iceland, MaCuMBA researchers not only networked and discussed their latest discoveries; they also undertook sampling expeditions to enlarge their collections of marine microbial specimens. PhD students and postdocs met and socialised at their own forum 'Young MaCuMBA' and exchanged project news, ideas, opinions and concerns.

On the look-out for future funding

The project members will hold their final conference, 'The Marine Microbiome – Discovery & Innovation' in Berlin, Germany, from June 27 to 30, 2016. The meeting is expected to bring academia and industry together. "So far, we do not have new pharmaceutical compounds or ready-to-use commercial products," said the project coordinator Stal. "A four-year project is too short to get from developing the techniques of isolation and cultivation of marine microorganisms to actual pharmaceutical and biotechnological applications. Unfortunately, the European Commission has no call in its Horizon 2020 Programme that would allow us to continue the project with European funding. We hope that in the near future, new funding opportunities will emerge."

Bettina Dupont

Peter Lawrence and science politics

The Last 50 Years

Science Mismanagement and Creeping Corruption

In his latest polemic on the parlous state of science, Peter Lawrence provides an overview of a half-century, in which science has been increasingly disrupted and distorted by "mismanagement" and "mismeasurement". Jeremy Garwood analyses the main points of his argument and some of the reactions to his claims.

Photo: Fotolia/Antonio Gravante

Peter Lawrence is a successful scientist, who, during his long and distinguished career, has been openly critical of the abuses and changes that have afflicted the system of scientific research (see text box 'A Half-Century in Science' on pg. 20). He was recently invited by the journal *Current Trends in Developmental Biology* (on the occasion of its 50th anniversary) to write an essay, in which he would look back over the past half-century of research.

Instead of concentrating upon his personal research interests, he chose to write about the negative changes that have affected all of science during this period. His essay, titled 'The last 50 years – mismeasurement and mismanagement are impeding scientific research', brings together views and critiques that he has been expressing and refining during the last two decades (text available at: *http://making-of-a-fly.me/files/pdf/Lawrence-2016.pdf*).

The end of science?

He has concentrated his analysis into seven specific changes that he says have proved "disastrous to the life of scientists and to science itself" (see text box 'A summary of the central claims' on pg. 22). Commenting on this essay, Tim Birkhead, professor of behaviour and evolution at the University of Sheffield, wrote that Peter Lawrence had "elegantly articulated" reasons why he and many of his colleagues now fear that matters have become so bad (at least in the UK) that we are approaching "an end to science", in which "our creativity and even our integrity is being suffocated by the modern bureaucracy and politics of research and teaching" (*Times Higher Education*, 24/03/2016).

Warped career motivations

Throughout his essay, Peter Lawrence illustrates his points with specific examples but here, we will concentrate on the central points. His first claim concerns the motivation of scientists for dedicating their lives to a career in scientific research. In 1962, the year when he himself first started to do research, it was not uncommon for scientists to be motivated by the goal of higher truth and discovery for mankind. There was an "atmosphere of excited curiosity among graduate students."

A half-century later, this spirit has been replaced by the "pragmatism" of career choice. Now, he finds it is relatively rare for students to discuss their projects in the coffee room. "Much more time is spent worrying about the next career step, about publication and the politics of science. This change has even affected my life; I now get more invitations to talk about scientific politics and careers than about my research." This sense of career insecurity has been driving many students away from science. "Knowing how tough, insecure and financially unrewarded the scientific life is, most bright students prefer other paths such as finance, law, and medicine," Lawrence says.

Suffocating bureaucracy

He denounces the asphyxiating effects of a huge increase in the bureaucracy and administration of science. Scientists are subjected to control by non-scientists who either don't understand or don't care about the objectives of research and researchers. Their research is assessed and controlled by people who have little understanding of the "summer lightning of creativity or the inconvenient truth that trying new things carries a big risk of failure." Because research is about investigating the unknown, we cannot predict what we will find.

Lawrence points at the impossible "fiction" of grant proposals that claim to be capable of mapping out in detail the future progress of research projects – how interesting is research that is so easily predicted in advance? In addition, grant applicants are now expected to promise economic deliverables from their research and to make knowledgeable statements about any future social and economic impact. With no facts from experiments that have not yet been performed, how can it fail to be fiction, albeit based on informed guesses that are adapted to please the grant committee in the hopes of winning the grant?

The last decades have seen an inexorable rise in university administration with suffocating effects on teaching and research, their "core purposes". Instead, new administrative posts claim to provide "human resources" and "research support". Science is giving way to what Peter Lawrence refers to as "meta-science". These are jobs in science administration for granting bodies and journal editors. It is now relatively easy to find qualified scientists to fill such posts, since meta-science appears to offer more job security and money than research itself.

Peter Lawrence also points to examples of how university administrators are setting artificial production targets for researchers. Previously, administrative staff were appointed to "facilitate" the work of academics, dealing with the paperwork and leaving researchers to get on with their research. Nowadays, the roles often seem to have been reversed. Scientists spend more and more time doing paperwork for the administration, working in the interests of the university's position in the "rankings". Who is there to serve whom? A recent analysis by the *Times Higher Education* confirmed that most UK universities now employ less academics than "support staff". On average, 43% of UK university employees are now managers, administrators and secretarial staff (*THE,* 3/09/15). To maintain their financial status, UK universities compete for government funds in research assessment exercises, like the Research Excellence Framework (described in 'Excellence or Non-sense' *LT* 5-2015 p.28-31).

Disastrous practices

By putting additional pressure on its academics, university managers seek to improve their "metrics", e.g. to achieve better research scores in REF 2014, Queen Mary University London sacked full-time lecturers in biology and medicine, who, it claimed, had failed to publish in journals with high-enough impact factors, or to win enough grant money ('Academic values no longer add up' *LT* 4-2012 p.20-25). In 2014, the suicide of Stefan Grimm at Imperial College London was also directly linked to "bullying" demands that he find more research grants (see interview with David Colquhoun 'Bibliometricians are the curse of the age' *LT* 6-2015 p.20-23).

Peter Lawrence says the transmission of scientific understanding has been harmed by changes in publication practices. These have transformed research papers into numerical measures of research output that can either make or break careers and research domains. Lawrence himself was an editor for over 30 years on several prominent scientific journals (notably *Development*, *Cell* and *EMBO Journal*). He says that the number of publications and the journal, in which they are published, are being used for "ranking" researchers and their research. Since this affects their careers and research funding, it has had the effect of pitting one researcher against another in a competitive contest with many losers, not least science itself. Lawrence says this is part of an increase in the "audit society" that has risen since the 1980s (that he previously denounced, e.g. in 'The Mismanagement of Science', *Current Biology* 2007, 17: R583-5).

Currently, there are "top" journals that have become like places of "pilgrimage" for

researchers. As a result, the original purpose of research publications – to communicate science – is less important for researchers than their need to be seen to publish in such journals. Furthermore, he notes that the power of citations has reinforced the position of these top journals because researchers tend to preferentially cite them over smaller journals in an effort to make their own papers seem more important.

Slaves to the system

The mismanagement of science has also "damaged" the practice of publication, i.e., how journals now operate and what they publish. He finds that a paper's main function in many journals is no longer scientific communication but rather "the badge of honour" because journals and their editors have themselves become slaves to impact factors. Meanwhile, scientists are now so obsessed by the impression they must publish in high-ranking journals that they "make less time for reading and comprehending because, in order to survive, they must put so much effort into writing manuscripts and manoeuvring them into journals."

Since journals favour "fashionable papers", e.g. with medical relevance, scientists have also adopted a "play-safe" attitude in their choice of projects. The result is that the "sense of adventure so vital to research has been dampened." This has further been exacerbated by the power of reviewers to make or break an article during the peer review process. He says that reviewers have acquired too much power over authors, whose "fear of antagonising reviewers has led to an increasing tendency to be diplomatic, to write manuscripts where conclusions are obfuscated and opinion is hidden". The value of review articles has also suffered because "most reviews simply repeat current dogma and fail to examine the evidence behind it."

A nightmarish experience

The funding system for research – the 'granting system' - is "dysfunctional". Getting money has become a nightmarish and constant rush to find and apply for grants. There are "different eligibility requirements, clashing deadlines, byzantine web-based forms" and time limits, in which everything has to be completed and submitted. Often, after all this time and effort, the awarded grants are for such short periods that the researchers are obliged to pass yet more time desperately looking for another grant that can follow on from it.

The combination of all these "political" changes over the last 50 years has impacted on research itself. Lawrence says it has resulted in "an insidious corruption of the practice of research." He points again to the example of research publications. The primary purpose of publication has changed from communication to producing "tokens" that will yield salaries and grants. As a consequence, the focus of research is now to produce enough papers to compete and survive. This has generated a sense of urgency to publish projects as soon as possible, often before they have become "fully rounded and completed stories." In such circumstances, it is not surprising that published results are increasingly non-reproducible. Furthermore, the lack of time to develop mature projects is discouraging researchers from exploring new ideas because they cannot "afford to do really adventurous research these days, it's just too risky."

Grant and reviewer tyranny over soon?

Nevertheless, Peter Lawrence ends on an optimistic note. He says there is a growing awareness of the problems that have arisen and attempts are being made to try to bring an end to bibliometrics and grant tyranny. For example, the Wellcome Trust

Peter Lawrence – A Half-Century in Science: Research, Publications and Politics

In fact, Peter Lawrence started his research career in 1962 – more than a half-century ago – at the Department of Zoology, Cambridge University. For his PhD, he studied pattern formation in insects, then did a postdoc for two years in the US, followed by two years back in Cambridge. He then "got recruited by Sydney Brenner and Francis Crick" to a permanent research position at the Medical Research Council's noted Laboratory of Molecular Biology in Cambridge. He remained there for 37 years until obliged by their "age discrimination" policy (he was 65) to set up a new, Wellcome Trust-funded, laboratory in the Department of Zoology, where he is still an active researcher. Peter Lawrence's work has defined the concepts of polarity, morphogenetic gradients and cellular compartments as key components in the growth and patterning of animals. He has been an editor of the journal *Development* for 33 years, and on the editorial boards of *Cell* and *EMBO Journal*. He is a member of EMBO, Fellow of the Royal Society and of the Royal Swedish Academy of Sciences.

From *Development* 143: 183-5

Despite his personal success in research, Peter Lawrence became increasingly aware of the difficulties that younger scientists around him were facing in a changing research environment. In the 1990s, he decided to speak out about what was happening, both in his research seminars and in a series of high-profile essays, e.g. 'Rank Injustice' (*Nature* 2002, 415:835-6), 'The politics of publication' (*Nature* 2003, 422:259-61), 'The mismeasurement of science' (*Current Biology* 2007, 17, R583–R585), and 'Real lives and white lies in the funding of scientific research: The granting system turns young scientists into bureaucrats and then betrays them' (*PLoS Biology* 2009, 7. e1000197). In a *Lab Times* interview in 2011, he discussed how he had become aware of the increasingly disastrous changes that were affecting science ('The heart of research is sick', *LT* 2-2011 p.24-31) and recently contributed to a series of articles at *LT* online – 'Is there "Institutionalised Corruption" in Science?'.

-JG-

has simplified the bureaucracy of its grant applications, placing more emphasis on a small number of the applicant's best research papers that can realistically be read and assessed by examiners. He says that publication initiatives, like the launch of the journal *eLife,* represent attempts to eliminate 'reviewer tyranny' over authors.

The founding editor of *eLife*, Randy Shekman, used his media prominence as a Nobel winner in 2013 to urge scientists to boycott 'luxury brand' journals. The movement around the 2013 'Declaration on Research Assessment' has also attempted to find alternatives to the grinding logic of impact factors ('SF DORA: Time to change how research is assessed', *LT* 5/2013 p.18-23).

In his conclusion, Peter Lawrence reasserts the fundamental importance of science for human civilisation since he insists "education that is founded on rational argument and scientific research must be the only feasible way to save our planet and our species." We need to resist tendencies to find "supernatural" explanations; science must be about understanding and innovation. "Our dependence on phoney measurements and bureaucracy must be abandoned and our processes must again reward originality and risk-taking."


Photo: Post-Polio Health International

Back in the old days (ca. 1960) , when smoking was still allowed in the lab. In this picture, Albert Sabin (right) works on the poliovirus vaccine.

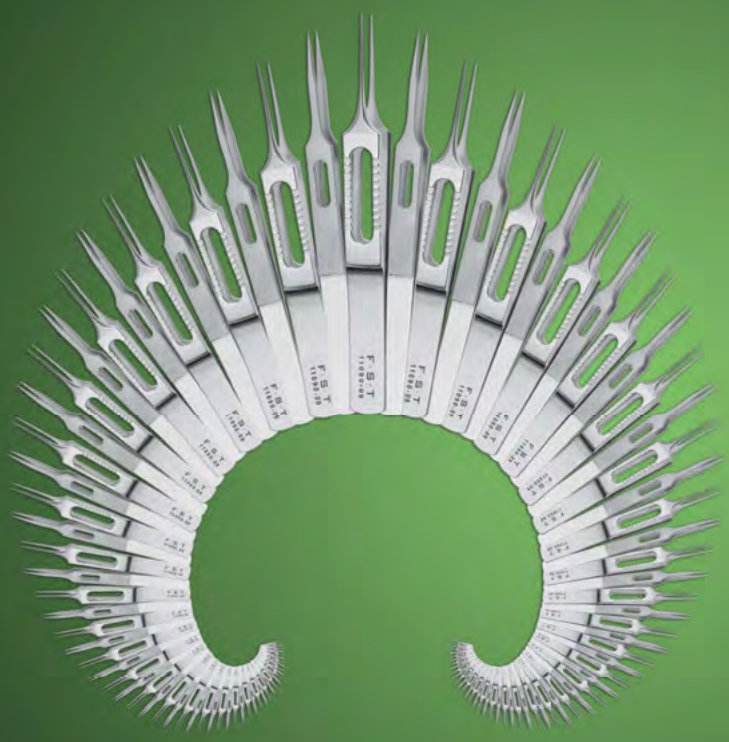
Reactions to Peter Lawrence's polemic

Peter Lawrence told *Lab Times* he had received a lot of supportive feedback but said it was possible that those in disagreement didn't write. "Anyway, the people who write seem to agree with me on everything, or almost everything and so I believe I have discovered a large constituency of scientists who have detected the same structural problems."

Commentators gave examples of bad administration, corrupt science, people giving-up research, discouraging new entrants, frustrations with the reviewing system, etc.

Roberto Oyarzun, Professor of Geology at the University of Madrid, noted that it is hard to get academics to think about these problems. "I've been discussing these topics with my colleagues for years, alas, a dialogue with a concrete wall would have been more effective (although a few are now starting to realise what's going on)." He suggested that this is similar to the "old story of the frog that is placed in boiling water:

F · S · T®
FINE SCIENCE TOOLS



WORK OF ART

Fine Science Tools is committed to serving the world's scientific and biomedical research communities with a full range of precision surgical and micro-surgical instruments. Unparalleled quality and customer service has made us the leading distributor of fine European surgical instruments worldwide.

FINE SURGICAL INSTRUMENTS FOR RESEARCH™
Visit us at **finescience.de** or call **++49 (0) 6221 90 50 50**

it will jump out, but if it is placed in cold water that is slowly heated, it will not perceive the danger and will be cooked to death."

Venkatraman Ramakrishnan at the LMB in Cambridge (and Nobel prize-winner in 2009) said he was very glad the essay had been written because "the scientific endeavour is increasingly a stressful and misdirected enterprise." He hoped the article would have some impact but feared that "scientists are so harried and have little power because our 'product' seldom affects the short term."

Inviting bullshit

"As usual, you hit the nail on the head," wrote Tim Hunt (another Nobel prize-winner in 2001). He particularly deplored 'impact statements'. "I've no objections to retrospective enquiries – what did you find out, and was it useful? But prospective ones are simply inviting bullshit."

Markus Noll at the University of Zurich said that senior scientists must speak out. "It is comforting to hear someone of your high scientific stature with whom I agree so much, while there are so few who dare to think as you do and with whom one could share the thoughts because they would only ridicule or ignore you. It is crucial that there are a few scientists like you who speak out clearly and argue based on known facts."

Is it a good idea for young students to enter scientific research? Given the problems he has described, Peter Lawrence honestly admits that he now tries to dissuade young people from a career in science. "They have to be obsessed to do it, in my opinion. It's a bit like going into poetry or acting, it is creative and can be rewarding but you spend all your time in great insecurity, fighting the administration that is supposed to help you. It is sad as we depend on scientific research for nearly everything, as I try to emphasise."

Several commentators lamented the waste of talent when promising researchers are forced to leave science for lack of opportunities. Alexandra Joyner at the Sloan Kettering Institute in New York wrote, "Every time I see another of our best and enthusiastic young scientists reject an academic career, I feel sad that the system has let them down." Walter Gratzer, emeritus professor at King's College London, said he doubted whether, "starting out now, I would choose to go into science. Perhaps I would aspire to a career in administration, or one of the other growth areas, and become a lab safety officer."

Better system, more good science

Adam Wilkins, managing editor of *BioEssays* from 1984-2008, made the point that despite the decay and dysfunction of the system described by Peter Lawrence, good science had still been produced. However, he said, this "raises the question of whether so much more might have been and could be accomplished with a much better functioning system."

Dan Hultmark at Sweden's Umea University, thinks science has been mis-selling itself. Good, basic science "answers important questions about the world around us: What is a living organism and how is it created? How does it work? Where do we come from? What is our place in the Universe? What, really, is mind and thought? These are existential questions, for which humans have always looked for answers, and they have been ready to invest much time and resources to get even very vague answers." He says "our mistake" is to use economic arguments to justify our research – with inflated claims about the likely usefulness of our results – rather than to stress "the fact that we contribute to important knowledge, or that it is enormously stimulating to do so."

Signs of change?

On the positive side, Dan Hultmark sees "many protests against the ideologies of New Public Management and micromanagement. Things will get better but it will take time."

However, Pedro Saavedra (a graduate student from Cambridge, now in the USA) said that although he knew people who agreed with what was said, "they wanted to hear some solutions/alternatives as well..."

A more radical interpretation was provided by France's Alain Trautmann. He told *Lab Times* that "many scientists are currently experiencing a deep unease" in the French research system and pointed to a similar analysis that he published last year "Malaise dans la Recherche" (in French, *http://tinyurl.com/jgb59ng*). Trautmann, an emeritus research director in France's CNRS, has been a prominent critic of French government policies for research and higher education. He was a spokesman for the movement 'Sauvons la Recherche', a co-founder of the protest movement 'Sciences en Marche' and co-author of the European protest 'They have chosen ignorance' (discussed in *LT* 3/2015, p.22-27).

While he agreed with the points made in Peter Lawrence's "remarkable" essay, he felt that it had not gone far enough in identifying the cause of the problems afflicting science. "I think that it is possible to go one step deeper in the political analysis of this situation. What has been the motivation of the politicians and administrators who have pushed the system in this direction?" He says many of these problems are a direct consequence of political changes that have introduced the agenda of New Pub-

Summary of the Central Claims

In the last 50 years, there have been many changes to the substance, conduct and style of research. Many of these changes have proved disastrous to the life of scientists and to science itself:

- the near-romantic spirit of adventure and exploration that inspired young scientists of my own and earlier generations has become tarnished
- now, many of us feel beleaguered by bureaucrats and politicians
- the core purposes of universities, teaching and research are being eroded by excessive administration
- the number and locations of our publications are counted up...and then used to rank us, one against another
- the granting system is so dysfunctional it could not have been designed
- mismeasurement has damaged the practice of publication itself
- there has been "an insidious corruption of the practice of research".

It is, therefore, crucial that the primary purposes of science to understand and to innovate can once again become the overt aims of researchers. To achieve this, our dependence on phoney measurements and bureaucracy must be abandoned; our processes must again reward originality and risk taking.

Source: Peter A. Lawrence "The last 50 years -- mismeasurement and mismanagement are impeding scientific research" *Current Topics in Developmental Biology,* 2106, 116: 617-31.

lic Management and Neoliberal economics throughout our societies. This has notably made systematic accountability a means of control – "every aspect of our lives is systematically measured, counted, and we spend an increasing fraction of our times making these measures and rankings. What cannot properly be measured (love, happiness, generosity, solidarity) counts for nothing. The important notion is profit, if possible short-term profit. Most citizens have suffered from this situation without understanding its power and its consequences."

Contrary to Peter Lawrence, who sees signs of change in a journal like *eLife*, Alain Trautmann says he is convinced that the situation of Higher Education and Research "will continue to worsen as long as our societies consider that the economy and profitability are the alpha and omega for the functioning of all sectors in society."

"None of these manifestations of the sickness of public research activity is specific to the Anglo-Saxon world." They also exist in France where the criterion of short-term profit has become a priority (which is absurd for basic research); an increasing mistrust of scientists by politicians has developed, and a whole administrative army has flourished for measuring and controlling research activity.


50 years later, the only smoke in the lab comes from cryopreservation devices. How times change.

Photo: United States Department of Agriculture

Politicians' ignorance

Trautmann says politicians have an "abysmal ignorance" of what research really is, as explained in the Open Letter 'They have chosen ignorance' that has gained support from researcher protest movements across Europe.

So, should we be looking at science's current problems as part of an overall change in how our societies are being organised? The scope of these changes has been described, for example, in George Monbiot's essay 'Neoliberalism – the ideology at the root of all our problems' (*The Guardian,* 15/4/2016). But then, as he writes, it is not enough to recognise what has been happening during the last half-century, because "a coherent alternative has to be proposed" that is "a conscious attempt to design a new system, tailored to the demands of the 21st century." A total rethink for the next half-century?

Jeremy Garwood

Dispensette® S
Bottletop dispenser

The new one!

BRAND

Dispensing safely and more convenient than ever!

Faster Priming

Less force needed
during dispensing

Volume selection with scalloped track

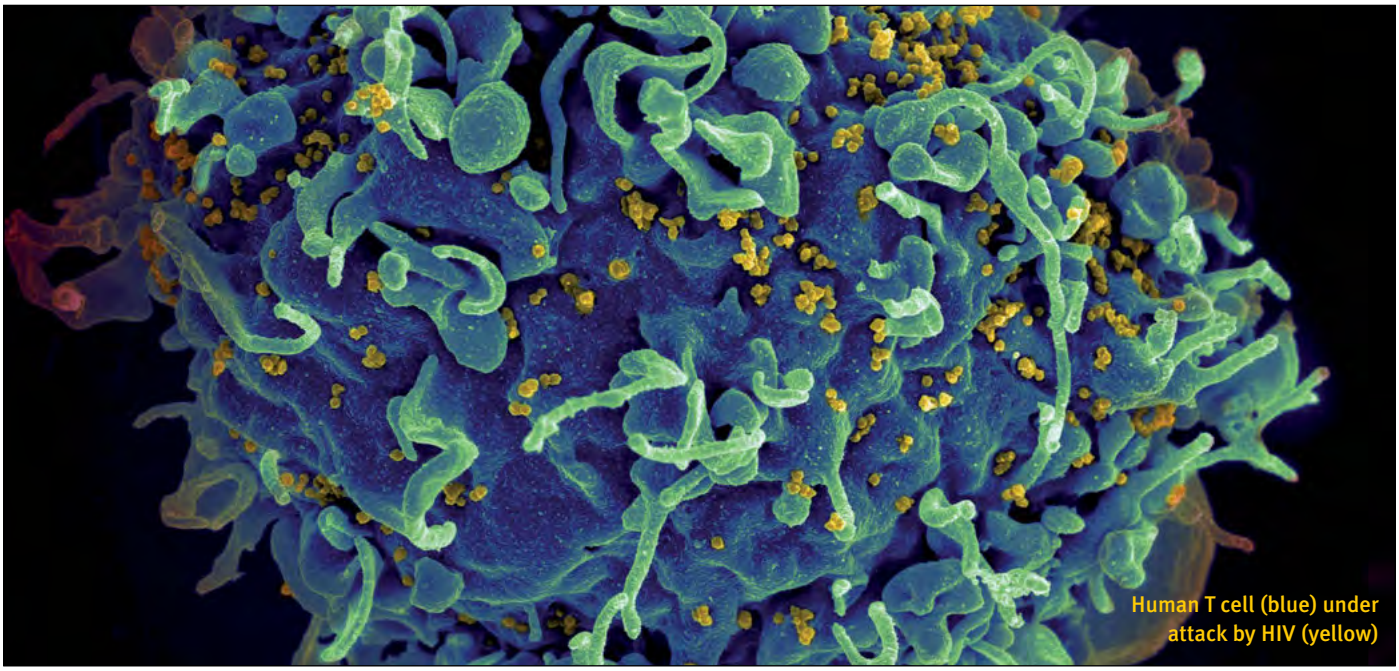
New discharge tubes
with or without recirculation valve

New valve system
no sealing rings necessary

New 1 ml size

BRAND GMBH + CO KG

P.O.Box 1155 · 97861 Wertheim · Tel.: +499342808-0 · info@brand.de · www.brand.de



Human T cell (blue) under attack by HIV (yellow)

Picture: National Institutes of Health

A conversation with Joachim Hauber, Hamburg

"Maybe in Two Years, We'll Be Able to Treat the First Patient"

HI viruses very cleverly "sneak" their own genes into the genome of infected immune cells, and just lay low, undetected and untouchable. For many years, a cure for AIDS has eluded scientists as it seemed impossible to dig back out the viral genes but these little infiltrators may have finally met their match.

Joachim Hauber is a German virologist striving to find a cure for AIDS. His perhaps most well-known work came in 2007, when he and his team successfully did what many believed impossible: they eliminated HIV DNA from infected cells by means of a cleverly concocted Tre recombinase. Recombinases are certainly nothing new and researchers have known for a long time how these "nifty" enzymes can cut and recombine specific parts of the DNA. The trick was to take advantage of this cut-and-paste ability to design a new recombinase, able to target a specific sequence present in the HI virus. In theory, this process can not only stop viral replication but, more importantly, it may actually eradicate the virus from infected cells.

For practical reasons, the original work only targeted a sequence present in a minority of HIV patients but Hauber didn't give up and has recently published a new study, expanding this approach to cover over 95% of patients. There's still a long way to go but this could become the alternative to the standard antiretroviral therapy. *Lab Times* went to interview Joachim Hauber to find out a little more about his work.

Lab Times: How did your interest in viruses start?

Hauber: It was a long time ago when I started looking into viruses, in 1983. Before that, during my PhD, I worked in a lab interested in retrotransposable elements in yeast. At that time, this was an interesting field and we discovered that the genome of yeast harbours transposable retrovirus-like elements, the so called Ty transposable elements. We published the first full length sequence of Ty elements, which was actually my first paper. I was lucky to be first author (*Nucleic Acids Res,* 13:2745-58). It was at a time when DNA sequencing was done by ourselves, as commercial options were not available.

After my thesis, I wanted to do a postdoc in the US and I joined Brian Curry's lab, who's a very successful HIV molecular biologist. He started his own lab at the Roche Institute of Molecular Biology, in Lincoln, New Jersey. It was very early for HIV research in Europe but in the US it was already picking up speed. At the time, I was his first postdoc. It was a very small lab; just Brian, myself and a lab technician. A few years later, I moved to the Howard Hughes

Pictures(3): HPI

Joachim Hauber studied biology at the Tübingen University and then did his PhD at the Ludwig-Maximilians University, Munich. After two postdoc positions in the US, at Hoffmann-La Roche in New Jersey and the Howard Hughes Medical Institute in North Carolina, he returned to Europe in 1988 to work at Sandoz' Research Institute, leading its molecular biology division. Later, he became professor of molecular virology at the University Erlangen-Nuremberg and finally, head of the Antiviral Strategies Unit of the Heinrich Pette Institute – Leibniz Institute for Experimental Virology in Hamburg.

Medical Institute, in Maryland, USA and eventually back to Europe. So, I've been working on HIV since 1986.

You've been involved with some crucial papers in your career. Which ones would you consider the most important and exciting?

Hauber: This is a really difficult question. I mean, I've been lucky I have been part of some important papers over my entire career. Once in a while you uncover something really important. During my postdoc times, certainly it was one paper in *Nature* and one in *Cell* dealing with the HIV-1 retrotransactivator protein. The first one, in 1988, described the target sequence in HIV-1 direct response element (*Nature,* 335:181-3) and the second one, in 1989 – I think maybe even more important – described the Rev protein transdominance mutant (*Cell,* 58:205-14). I think it was an important paper, not only interesting for the people in the field but it also triggered the idea of transdominance division in respect of HIV replication.

"I've been lucky I have been part of some important papers over my entire career."

Later on, I moved back to Europe, originally to the Sandoz Research Institute in Vienna, Austria, which later became Novartis. Sandoz/Novartis also used our model into the 1990s to develop therapies for HIV disease. At that time, there was no good antiretroviral therapy, as combination of therapies only started in the mid-1990s. At the beginning of the 1990s – given that there was no good therapy available – Sandoz decided to use this model system for managing therapy. I think this is what also makes these papers important, in a broader sense.

"The technology behind tailored recombinases was developed in a time before zinc-finger nucleases and CRISPR/Cas9."

Later on, in 2007, it was our first study in *Science*, on Tre-recombinase or the first tailored engineered recombinase able to excise the HIV genome from infected cells (316:1912-5). I think this was also a landmark from the technology point of view. The technology behind tailored recombinases was developed in a time before zinc-finger nucleases, CRISPR/Cas9 and so on. It was completely unexpected that it would be possible to reverse an HIV infection at the molecular level.

The 2007 paper in Science *was hailed as a potential cure for HIV...*

Hauber: Basically, the 2007 paper was a proof-of-principle study showing that it's possible. The recombinase we used at that time, for technical and historical reasons, only targeted very rare sub-type A isolates. With that recombinase, we were able to show that it's possible to remove the infection but with respect to treating patients, this Tre-recombinase can only treat less than 1% of all HIV patients. This means it's not useful for translational development or application in the clinic. At that stage, we didn't know if we would succeed,

but we knew we had to develop something with broader anti-viral activity.

How did you come up with the idea of using recombinases anyway?

Hauber: Our work on recombinases is based on a collaboration with the lab of Frank Buchholz [originally located at the Max-Planck-Institute of Molecular Cell Biology and Genetics in Dresden, now at the Technical University Dresden]. Buchholz is an internationally renowned expert on designer-recombinases/enzymes. Around 2006, we started the project, which continues to this day, to use tailored recombinases to excise the integrated HIV proviral DNA from infected cells. This was based on the idea that only removing integrated HIV in patients will allow a potential HIV cure (functional or sterilising).

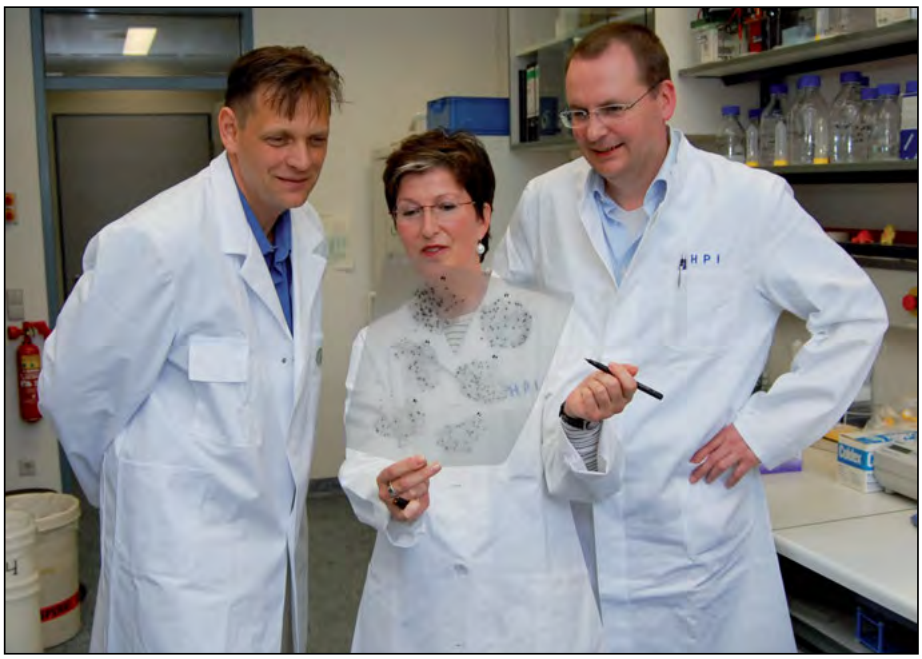
We believe that provirus excision will be an essential component of any future cure approach. Thus, no single approach will probably work. Instead, any successful cure (if feasible at all) will require the combination of various approaches, such as provirus excision, advanced immune therapies (vaccination), next-generation cART, etc.

So, these recombinases recognise a specific target?

Hauber: Recombinases are nothing new, they exist in bacteria, yeast and so on. They have their target sequences and, if these target sequences are present, they will scan the genome to find them. The process, in our view, can be compared to editing with zinc-finger, TALENs or CRISPR/Cas9. These enzymes do the excision, which we are interested in – in our case, the HIV genes – in an extremely accurate and precise manner. These recombinases do not depend on cellular pathways, they do it by themselves and will not create free DNA. So it's absolutely predictable what the result will be.

"It was completely unexpected that it would be possible to reverse an HIV infection at the molecular level."

The trick was to mutate a recombinase. Look at the native recombinase target sequence [called LoxP, and widely used in molecular biology to generate transgenic mice] and you will never find a similar sequence in any HIV isolate, so the trick was to mutate the original recombinase to recognise a completely different sequence in the HIV genome.



Inner-German collaboration at its best: Frank Buchholz, (left, Technical University Dresden) and the Haubers, Joachim with his wife, Ilona (Heinrich Pette Institute, Hamburg).

How difficult was it to develop this procedure?

Hauber: In the beginning it was very difficult. Many labs failed to mutate existing recombinases, such as Cre, in a way that the product recognises native HIV sequences. The reason for that is that the 34 bp native target of Cre [i.e. the loxP site] is a symmetric sequence. Unfortunately, HIV only contains potential target sequences that are asymmetric with little resemblance to loxP. This problem was solved by Frank Buchholz's idea to generate recombinase-intermediates that each recognise only a subregion of the final HIV target sequence. These intermediates were then combined by gene shuffling, resulting in the final designer-recombinase, i.e. Tre-recombinase that now recognised a native LTR sequence, termed loxLTR.

"Recombinases do not depend on cellular pathways and will not create free DNA. So it's absolutely predictable what the result will be. "

You now have a new manuscript in Nature Biotechnology *with the potential to become another landmark paper. How does this move forward from previous work?*

Hauber: From my point of view, the most interesting aspect is that we were able to generate another tailored recombinase but this time, able to recognise and target the vast majority of all clinical HIV isolates. Overall, more than 90% efficient, more like 96-97%. I'm sure that before we published this work, many HIV experts never expected something like this would be possible because there was always the notion that HIV mutates so fast, impeding any treatment. However, succeeding to identify highly conserved target sequences that nobody expected to be there is an important contribution, which will open the door for clinical studies using this target sequence.

While Tre was good for proof-of-concept studies (*Plos Pathogens,* 9:e1003587), the caveat was that Tre only recognises a rare HIV subtype A isolate. For broad clinical testing/application, however, it is desirable to particularly treat HIV subtype B isolates. This problem was solved by engineering Brec1 (Broad range recombinase 1), which targets >90% of all known HIV-1 clinical isolates, particularly >94% of all known subtype B viruses (*Nat Biotech*, 34:401).

What exactly is the new genetically engineered Brec1-recombinase targeting?

Hauber: Brec1 recognises a 34 bp sequence in the R-region of the HIV-1 LTR, termed loxBTR, or lox Brec Target Region. The loxBTR sequence is highly conserved among clinical HIV-1 isolates. Upon recognition of loxBTR, Brec1 recombines both target sites, located in the sequence-identical 5'- and 3'-

LTR, and thereby excises the intervening sequences, the HIV genes, in an "error-free" manner. The latter ensures highly accurate and precise provirus excision, which is of utmost importance with respect to any future clinical application. The excision product cannot re-integrate and is degraded over time by cellular nucleases. The formerly infected host cell is now free of HIV.

Given these positive results, you must be planning clinical trials already! How long before you think you can treat your first patient?

Hauber: First of all, we've now finished all pre-clinical studies as far as possible. For clinical studies we need approval by an overseeing agency, which in Germany is the Paul Ehrlich Institute, near Frankfurt. It's clear what we have to do but the problem is finance. Because we have to do gene cell therapy, all the reagents have to be produced under Good Manufacturer Practice conditions. As a research institute, we cannot do it ourselves. We need a supplier but there are not many in Europe and the production process is really expensive.

We're looking for potential investors to finance that study and as soon as financing is secured, we can move on. This will not mean that the next day we'll be able to treat patients. If everything works out and we're able to secure financing, maybe two years from now we'll be able to treat the first patient.

This will also be a very small study, ten patients, and we are not able to treat regular HIV patients in that first study. This is due to the ethical considerations, as for these patients there is a good therapy already, such as retroviral therapy, so they can deal with their infection pretty well. These patients should not be exposed to a brand new gene therapy with unknown risks. What we can do in the first study is to treat patients that suffer from another life-threatening condition, such as cancer, and are also HIV-positive. In this context, this additional treatment for HIV could be acceptable for these patients, as they have to undergo chemotherapy for the tumour.

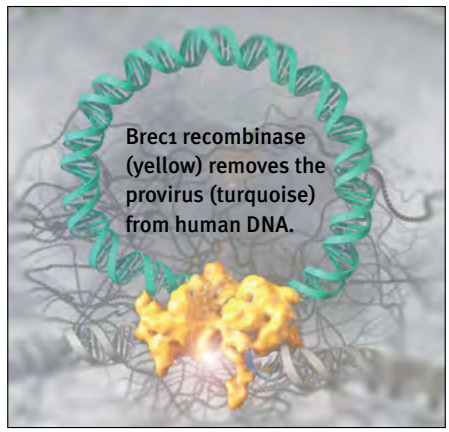
In the midst of developing ways to cure HIV, you're also organising the 6th European Congress of Virology (ECV2016) next October in Hamburg, Germany. Do you have a main theme for this conference?

Hauber: We aim to cover every aspect of virology. We want to present state-of-the-art basic research but also want to bring in clinicians. As a result of my own experience over the last years, I know we get the best results when basic researchers and clinicians work closely. Out of basic research can originate new targets and new ideas. We will basically have everything from basic to clinical virology and hopefully have fruitful discussions and state-of-the-art lectures on new developments and what's going on in the field. We're expecting about 1,500 participants.

"We're looking for potential investors to finance that study and as soon as financing is secured, we can move on."

What made you decide to organise this congress?

Hauber: The official organising society is the European Society of Virology and I was nicely asked whether I would like to organise this congress. In a moment of weakness, I said yes!



Brec1 recombinase (yellow) removes the provirus (turquoise) from human DNA.

Recently, two major viral outbreaks have featured heavily in the news: the Ebola outbreak in Africa and the Zika virus in South America. Will these cases also be discussed at the congress?

Hauber: We will certainly have sessions of pathogenic and highly pathogenic viral infections. We'll have presentations from top people in the field dealing with Zika or Ebola, for example, so every aspect will be covered. Highlights will be, for example, sessions on "HIV and Hepatitis Virus Cure Approaches", on "Advanced Virus Diagnosis", such as ddPCR, or on "Emerging Zoonoses" like Zika.

"Any successful cure will require the combination of various approaches, such as provirus excision, advanced immune therapies, next-generation cART, etc."

Looking further afield and following the Zika outbreak, most major publications now seem to come from US scientists. It was the same with the Ebola crisis a few months ago. Why do you think European researchers are so slow to respond?

Hauber: My personal impression is that these outbreaks were just not on the European radar. Upon recognition, major research efforts are currently being undertaken in these fields and we will certainly see more data coming out of European labs in the near future.

Looking into the future, what do you believe are the most important questions to answer?

Hauber: In HIV, certainly the big question is whether we can achieve a cure and a cure that means getting totally rid of HIV. We are hopeful that with new therapies, like our approach, in combination with other therapies, such as interference with receptor usage, we'll be able to achieve what we could call a functional cure over the next ten years.

This is where, after a single treatment, the patient will be able to control the virus without additional drugs for a long time. The antiretroviral therapy is a big medical success but still there are downsides to it. The longer you take it, the more problems develop. If you consider that most people get infected in their 20s and you have to take medication for the rest of your life, it's not looking so good. I think we have to push this towards a cure. Whether we'll be able to achieve a sterilised [complete eradication of all traces of HIV] cure we don't know, but we'll keep trying.

In terms of research in your own group, what are your plans for the future?

Hauber: We're interested in developing novel and experimental therapies. Our translational research focusses on the clinical development – phase Ib/IIa studies in a small cohort of HIV patients – of Brec1 for HIV-1 cure. On the other hand, we are also looking more and more, experimentally and scientifically, into how HIV reservoirs are regulated. We really want to look at their biology and experiment with ways to interfere with these reservoirs. The lab is tending again toward more basic research, focussing on how viral (HIV) latency is established, maintained and regulated.

Interview: Alex Reis

Photo: www.publicdomainpictures.net/George Hodan

Transporter biology in Copenhagen, Denmark

Flickering Pumps

Despite their important functions in basic cellular processes, not much is known about ATPases' actual modes of operation. During the last ten years, Dimitrios Stamou has developed a new method to finally see the pump in action.

Certain proteins in cell membranes are the equivalent of bouncers in nightclubs and bars: they decide what is safe to go inside the cell and what is to be refused entry. Transporting anything from large molecules to ions, carriers come in many shapes and sizes.

While one type in particular – ion channels – has received a tremendous amount of attention after the patch clamp technique was developed in the late 1970s and early 80s, active transporters like ATPases have been the "poor cousins" in the family. Until now, it was actually impossible to detect any changes in these carriers, but a team based at the University of Copenhagen, Denmark, was set on developing a method to capture these elusive transporters in action.

Ten years in the making

Team leader Dimitrios Stamou has had this idea in his head for over ten years: recreate natural conditions in a Petri dish by means of small lipid vesicles, customised with specific transporters and anchored to the bottom of the dish with a line long enough not to cause any interference. The method sounds simple but the potential of these tiny balloons is enormous. "Because the volumes are so small, it allows us to actually record very minute amounts of solutes added into the little balloons," says Stamou. "Because the volume is so small, you could put in one or two molecules and the concentrations become very big." This aspect is crucial; as small volumes mean high sensitivity to detect small currents generated by these transporters.

Being able to "blow up" these balloons without bursting them was only the first hurdle. Next, there was the issue of how to measure these small electrical currents. It soon became obvious the method could not rely on direct electrical measurements like patch clamp. These transporters are a million times slower than channels, so picking up their electrical signal would be impossible. In fact, as Stamou explains, detection needs to be 100 thousand times below the limits of patch clamp. "Patch clamp measures electrical current but that was nowhere near, so we needed a different method, to gain a 100 thousand to 1 million better sensitivity."

The solution? Tag vesicles with the lipid-conjugated pH-sensitive fluorophore pHrodo and rely on fluorescence microscopy to measure proton movement. Total internal reflection fluorescence (TIRF) microscopy is sensitive enough to detect changes in pH inside the vesicle and catch the transporter in the act of carrying a "passenger". Incredibly, this approach allowed the researchers to look at individual transporters located in individual vesicles, resulting in an unprecedented level of detail about their activity (*Science*, 351:1469-73). "This is very handy because it allows us to monitor the same liposome, and thus the same transporter, for long periods," says first author Salome Veshaguri.

Photo: Stamou lab

Pumping up knowledge: Dimitrios Stamou

It's on. It's off. Now it's leaking.

For their first attempt using this method, the team decided to look at eukaryotic primary active transporter P-type ATPase, aka proton pump. Once the pumps were activated with Mg^{2+} and ATP, the measurements started coming in with the expected pH changes as protons were pumped into the vesicles. The overall picture looked good but soon, the researchers realised there was something lurking underneath these results. For sure, there was a pH gradient forming but, at closer inspection, not all pumps had joined the crowd. In fact, some vesicles didn't show a pH change at all, suggesting that their pumps were not active. It turns out that, against what had been firmly believed until now, these proton pumps are not actually active all the time but go through random cycles of active and inactive periods.

Curious about this on/off switch, the team looked at the profile of individual vesicles. Surprisingly, a model, including factors known to affect these pumps, wasn't a particularly good fit. In fact, the authors noticed the model was consistently underestimating losses due to leaks. In true detective style, the team analysed all their evidence and plotted together data from each vesicle. The results in the histogram were clear: two separate peaks meant two separate types of leaks. One of them wasn't exactly a surprise, as passive losses through the membrane are common knowledge. The other one, however, was not expected and it took some digging to find its origin.

In the end, adding ATPase inhibitor vanadate offered the solution: as this inhibitor blocked the pump, it also stopped the leaks. This showed these losses were not regulated by the membrane but by the pump it-

self. Veshaguri compared this leak to filling a swimming pool with a faulty pump. Most likely, "if, once in a while, this pump allows backflow of the water, it would be labelled as faulty and returned to the store," jokes the researcher. It goes without saying that re-plotting the graph adding the pump-dependent leak significantly improved the model. However, surprises still kept coming and the model had one final unexpected result to share: it would be easy to assume a random leak when the pump was working, but it turned out the pump only leaked when it was inactive. Going back to the swimming pool analogy, it would be like having a pump that leaks when it's switched off, allowing water to flow out of the pool.

Stamou emphasises how important it is to know when the pump is leaking. "Mechanistically, if you want to prevent or modulate [losses], you need to be able to measure the different types of leaks. If you know that your pump becomes leaky when you stop it, you try never to stop it. You try to keep it running all the time but you can do that only if you know what the problem is."

Control buttons

Once they knew the pumps could turn themselves on and off, the team was curious to find out what could be controlling this switch. The obvious option was to look at the regulatory domain (also known as R domain) by comparing the activity of the proton pump with and without it.

Adding justice to its name, the regulatory domain limited transport rates but, somewhat surprisingly for a domain considered to be auto-inhibitory, it actually increased the total time the pump was active and reduced losses due to leaks. It was actually easier to find an active pump in the ATPase version with the R domain than in the one lacking this section. "It was very important when we found out that the regulatory domain not only affects efficiency but also working hours," says Stamou.

Not surprising for a proton transporter, pump activity is also regulated by pH gradients established across the membrane. Exaggerated pH differences decreased the amount of time the pumps stayed active and increased the risk of leaks but, in this case, somewhat counterintuitively, it didn't affect the rate of proton transport when the pump was active. It's interesting to note that the transporters re-adjusted to an excessive pH gradient by switching off and allowing more leaks, rather than slowing down intake.

The team was quick to see the potential of these 'buttons'. "We found out that a break can be applied by affecting working hours but also we found out, very surprisingly, that another way to reduce the overall activity is by introducing a leak into the system," says Stamou. "Understanding this novel mechanism and developing tools to alter the time spent in any state, will be key to new approaches in drug development," adds Veshaguri.

Spread the method

Stamou's main wish is to spread the technique to other labs, working on transporter biology. "The first thing I would hope is that the method slowly becomes more accessible to other people. I would be immensely happy if this happens in five to six years from now," says the researcher.

In the meantime, from a research point of view, Stamou plans to tackle two problems. First, the next goal is to test different families of transporters to see if this on/off mechanism can be generalised. After all, it's also present in ion channels and there is no reason to expect it not to happen in other types of carrier. "We know ion channel fluctuations are present across all species, so, now that we found the transporters also fluctuate, you would expect it to be the same. But we don't really know and that's one of the things we're going to have to find out," says Stamou.

The second goal may prove to be harder to achieve, as the team wants to move away from using purified protein. The process now involves elaborate and time-consuming multi-step protein extraction and purification, before being mixed with a very specific lipid component to make these small artificial vesicles. For a new protein, this can take anything between one and ten years. "If we want the technology to become more popular, it will be important to remove this bottleneck. So we need ways to do this measurement, avoiding the purification and reconstitution process. There are a lot of ideas and we're working in this direction but this will take a little more time," says Stamou.

Mirroring what happened with the patch clamp technology, Stamou and Veshaguri really believe this method could grow bigger than its niche applications in the lab and actually leave its mark in the real world, by helping in the development of new therapeutic targets. "My greatest aspiration is that this method is going to have a big impact on transporter biology just like patch clamp had for ion channel biology," concludes Stamou.

Alex Reis

Occupational safety by ROTH
Dare to look!

• A wide range of products for safety and protection in the laboratory – goggles for everyone
• As a pioneer in the field of occupational safety, we offer decades of experience
• Highest quality & personal advice from an expert
• Extremely short delivery times
• Fair prices for the highest quality
We are the specialists for laboratory equipment, chemicals and life science.
Order on:
Tel. (+49)721/5606-0
www.carlroth.com
ROTH

Zika virus in Hamburg, Germany

Risky Travels?

Photo: Pixabay/stevepb

Fear of the Zika virus is spreading around the globe. Most scientists focus on the virus itself or its vector, the *Aedes* mosquitos. Dennis Tappe, however, had a closer look at immununological aspects of the infection and discovered that the majority of Zika patients has little to fear.

The spectre of Zika infection has been of grave concern to travellers to the Americas, Asia, Africa and the Pacific since the outbreak first began to gain publicity at the end of last year. Zika virus was first observed in a rhesus monkey in the Zika forest of Uganda, and the first human case was reported in 1952. The first large human outbreak occurred in 2007 in Micronesia's Yap Island. Since then, active transmission of the virus has been detected in the Pacific Islands and in South and Central America. Brazil reported a Zika virus epidemic early in 2015.

In April of this year, the World Health Organization (WHO)'s regional office for the Americas issued an alert, confirming that the virus is a cause of microcephaly, a condition wherein an infant is either born with an unusually small head, or the head stops growing after birth. The condition is often associated with severe developmental disabilities. Pregnant women have been advised not to travel to regions where Zika transmission is active.

Photo: D. Tappe

It's a mild disease without shock symptoms, says Dennis Tappe.

Zika causing muscle weakness

WHO has also confirmed the Zika virus as a cause of Guillain-Barré syndrome, a neurological disorder whereby the body's immune system attacks the peripheral nervous system. Patients, who develop Guillain-Barré syndrome, may experience muscle weakness, nerve damage and, in some cases, paralysis. Both of these conditions are extremely distressing but how much does an average traveller, who is not pregnant, need to worry?

Two different types of mosquitoes, *Aedes aegypti* and *Aedes albopictus*, transmit Zika virus. Both species are also responsible for spreading other viruses, such as dengue and chikungunya. The diseases are spread when a mosquito carrying the virus from the blood of an infected host bites a new victim, passing the virus into his or her bloodstream. There is additional evidence that Zika virus can be transmitted sexually between humans in semen and through blood transfusions.

Although an outbreak of Zika virus infection has been shown in some areas to increase the prevalence of Guillain-Barré syndrome in a population as much as 20-fold, the condition is extremely rare to begin with. The Centers for Disease Control and Prevention (CDC) estimates one to two cases per 100,000 people in the US. Regarding microcephaly, CDC estimates the condition is present in between one and 13% of pregnancies if Zika virus transmission occurs in the first trimester. This wide range points to how difficult infection with the virus can be to pin down.

For those who do not develop complications from the virus, Zika disease symptoms can be mild or even non-existent. A typical case of Zika virus infection without complications may include a mild fever, skin rash, and aching in the muscles, joints and head. All symptoms typically disappear in under a week.

Infected travellers

Researchers at the Bernhard Nocht Institute for Tropical Medicine in Hamburg, Germany, led by Dennis Tappe, set out to examine the immunology of Zika virus in a small group of otherwise healthy travellers returning from Asia and South America, who had become infected while abroad. They conducted a pilot study by analysing protein factors in the patients' blood at various stages of infection.

"We wanted to see what happens in patients who have a mild form of the disease, who are not pregnant and who are not suffering from Guillain-Barré syndrome or anything else," says Tappe. "This is a study on how the immune system works during the viral infection." Their paper, entitled "Cytokine kinetics of Zika virus-infected patients from acute reconvalescent phase", appeared in the June issue of *Medical Microbiology and Immunology (205(3):269-73)*.

As the sample size of this particular study was small, the conclusions that can be drawn from it are likewise limited.

Since the study was conducted in Germany, where Zika virus is not being actively transmitted, the researchers had to rely on returning travellers. As of yet, no large-scale study has been conducted that examines the effects of Zika virus on the immune system.

Zika virus is part of a family of viruses, known as flaviviruses that are commonly spread by mosquitoes and ticks. Its symptoms are similar to those caused by dengue virus, a different flavivirus with the potential for severe complications, and chikungunya virus, an alphavirus from a different family.

In order to examine how Zika virus interacts with the human immune system, the virus researchers examined serum from six different patients. They obtained a second sample from a few of the patients, for a total of nine samples. As the starting point of their analysis, they used the date of symptom onset, since the incubation period for the virus can vary between five and ten days, making it impossible to pin down when the infection first began. By measuring different immune parameters, the researchers caught a glimpse of how the body may grapple with the infection over time.

Cytokines are small signalling proteins released by immune cells. Their levels peak during an infection, an event the researchers describe as polyfunctional immune activation. Looking at cytokine levels in the returning travellers, the researchers observed immune activation and restoration consistent with a common virus. "What we see is that many, many cytokines go up in the acute phase and they go back down again in the recovery phase," says Tappe. "That's basically what happens when you get sick: you have a fever, you don't feel well and, after a while, you recover. This is what we see in the blood as well."

Everything normal?

These results were consistent with symptoms observed in patients who contract Zika virus without one of the complications. "We were not astonished by the results, as they confirmed the clinical observations that people are mildly sick and recover fully afterwards," Tappe says.

"This sounds pretty much normal. However," he adds, "we know from dengue fever cases that they have very high levels of these cytokines that sometimes do not return to normal. In those cases, you do not see immune reconstitution but rather aggravation." In other words, different viruses can cause cytokine levels to remain high, a condition known as cytokine storm. These cases are characterised by chronic inflammation and can develop into shock syndrome or haemorrhagic fever.

"This is something we do not see in Zika fever patients," Tappe says. "Things go back to normal and people feel healthy after a couple of days. These findings reflect a mild disease without shock symptoms." Although it is too early to draw any major conclusions about Zika virus immunology, the study's results are consistent with the observable symptoms, which typically disappear after a few days if they appear at all.

If Zika fever is by-and-large such a relatively mild condition, what makes some people susceptible to the debilitating effects of Guillain-Barré syndrome? "It is immune-mediated, so it depends on how your immune system is individually structured," suggests Tappe. "It could be a genetic factor that somehow primes your immune system on how to react to certain pathogens."

Zika virus is one of many pathogens linked with the syndrome. HIV, the flu virus and some bacteria that cause food-borne illnesses can also be a cause. Tappe likens the susceptibility to Guillain-Barré to that of allergies: not everyone who is exposed to these pathogens will react by developing the syndrome but certain individuals are predisposed to do so. Although Guillain-Barré has the potential to be fatal, even in the most severe cases, the majority of patients recover fully.

Not enough patients

In order to obtain more concrete answers about a condition that is largely still shrouded in mystery, larger-scale studies will be essential. Conducting such a study in Europe seems out of the question: "We do have more patients now but this would only double or triple the number," says Tappe. "That's not enough."

An area such as Brazil, where the Zika virus outbreak is ongoing, would provide enough patients for a thorough investigation. According to Tappe, such a study would involve three groups – patients without complications, pregnant women and patients who develop Guillain-Barré syndrome – containing several hundred people each. The immunological results would all be compared and these results might shed some light on what is significant and typical for Zika virus infection.

Only when such a study is completed, will we be closer to understanding and perhaps stopping the virus in its tracks.

Alexandra Taylor

Lab Times

Founded 2006. Issue 3, 2016
Lab Times is published bimonthly

ISSN: 1864-2381

Publisher:
LJ-Verlag GmbH & Co. KG

Office:
Merzhauser Str. 177, 79100 Freiburg,
Germany, Phone +49(0)761-286869,
Fax +49(0)761-35738

Management:
Kai Herfort, Tel: +49 (0)761-286869

Editors:
Ralf Neumann (Editor-in-chief), Kathleen Gransalke, Kai Herfort, Winfried Koeppelle, Harald Zähringer
Phone +49(0)761-2925884, *editors@labtimes.org*

Reporters:
Latika Bhonsle, Steven Buckingham, Florian Fisch, Jeremy Garwood, Karin Hollricher, Madhuvanthi Kannan, Karin Lauschke, Alejandra Manjarrez, Rosemarie Marchan, Alex Reis, Ralf Schreck

Graphics, Design and Production:
Ulrich Sillmann (Art Director), Kathleen Gransalke, Kai Herfort, Winfried Koeppelle, Ralf Neumann, Harald Zähringer

Cover Photo:
Jan Bommes (http://arcanumphoto.blogspot.de),
Editing: Kai Herfort

Sales:
Advertising Manager: Bernd Beutel
Top-Ad Bernd Beutel, Schlossergäßchen 10,
69469 Weinheim, Germany
Phone: +49(0)6201-29092-0
Fax: +49(0)6201-29092-20
info@top-ad-online.de

Recruitment:
Ulrich Sillmann, Phone +49 (0)761-2925885,
jobs@labtimes.org

Printed at:
Stürtz GmbH, Alfred-Nobel-Straße 33,
97080 Würzburg, Germany

Web:
www.labtimes.org, Webmaster: Carsten Rees,
Tel.: +49 (0)761-1563461,
webmaster@labtimes.org

Prices & Subscription rates:
- price per issue: €4.90
- research institutes/units: free of charge
- annual subscriptions for companies and personal subscribers: €27.-

Subscribe at *http://www.labtimes.org/kontakt/sub.html*, or mail to: *subscriptions@labtimes.org*

Bank Account:
- Volksbank Freiburg
KTO: 319 0 315; BLZ: 680 900 00;
BIC: GENODE61FR1;
IBAN: DE24 6809 0000 0003 1903 15

Evolutionary biomechanics in Liverpool, UK

Walk of the Titans

Photo: Lida Xing/University of Geosciences, Beijing

Think big. Now, put your thought on four legs and you'll have a sauropod, probably the largest animals that will ever walk on the face of the earth. The most interesting mystery about sauropods is how they adapted their giant bodies for locomotion. Vivian Allen and Karl Bates have an idea.

It's not very hard to find a child that is interested in dinosaurs and, equally so, an adult, who is fascinated to the point of actually studying palaeobiology. Some of those dinosaur lovers end up at the University of Liverpool, turning their fascination into scientific models. Karl Bates, at the University's Department of Musculoskeletal Biology, and first author of a recent paper on the evolution of the sauropod body plan (*R Soc Open Sci*;3(3):150636), has been fascinated with predatory dinosaurs since his master thesis. "The engineering challenges that dinosaurs of large size faced, to locomote safely and efficiently, sparked my interest in functional anatomy and biomechanics," explains Bates.

Vivian Allen, the study's last author, wrote his master thesis under the supervision of Michael Benton, one of the scientists that contributed to the popular series "Walking with dinosaurs". The show depicted the animals not as the monsters you know from blockbuster movies, but rather as normal life forms that "wanted to sleep, mate and eat things without getting hurt", Allen points out. Both Bates and Allen consider that the sauropods during the Cretaceous experienced one of the most fascinating processes in the evolutionary history of dinosaurs. From the middle Triassic until the late Jurassic period, sauropods evolved into the largest land animals ever.

Bigger than a German mining machine

By comparison, the largest land vehicle humans ever created is the German crane-looking Bagger 293. This enormous machine weighs 14,200 tonnes and requires five human brains to operate. Some sauropods, like *Diplodocus*, weighed on average 40 tons and had a comparatively little brain. There's no way, however, we can make Bagger 293 walk on four legs, whereas *Diplodocus* was chilling around using quadrupedal locomotion. How did they do it, biomechanically?

The initial input information for palaeobiologists usually comes from people like Mary Anning, who dug up skeletons from the crust of the earth in the 19th century. The probability of finding a complete skeleton is, however, as unlikely as winning the Eurojackpot. Tim Hunt once said that in science "you have to enjoy swimming in the sea of unknowingness". For Allen and Bates, that sea is composed of partially complete skeletons. This "uncertainty is just something you have to accept and present clearly in the results and conclusions when working with extinct animals", explains Bates.

Photos: V. Allen, K. Bates

Vivian Allen (left) and Karl Bates (right) think big – big dinosaurs.

Just imagine that, millions of years in the future, a piece of your pelvic girdle, half of your femur and a couple of your vertebrae, are found. Do you think those few bones are enough to reconstruct your appearance? Bates and colleagues approached this challenge by scanning the dinosaur bones and then creating three-dimensional models of the skeletons. The techniques used, included long-range laser scanning, digital photogrammetry and computed tomography scanning. The 3D models of bones do not, however, tell how much flesh was attached to them.

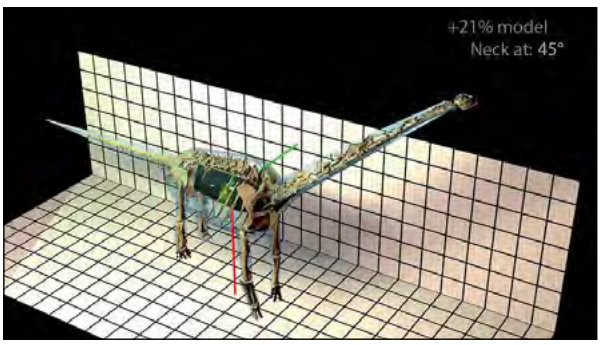
Imaginative humans

An unbiased reconstruction of a dinosaur body is hard, especially for humans with good imagination. Some time ago, the research team used animation software to


Picture: ДиБгд

subjectively recreate bodies based on the outline of some skeletons. It turned out that almost every scientist came up with a different picture! Apparently, using human brains to do image processing is too subjective and thus, Bates and colleagues decided to recruit silicon brains to do the job.

The "convex hull algorithm" was one of the most helpful lines of code. It calculates the minimal plane that results, when connecting dots distributed over a surface. If there are four points on a white piece of paper, the convex hull will be the square that tightly wraps around the dots. By digitising some vertebrae, the algorithm does a similar calculation but this time over the surface of the three-dimensional virtual bone. According to Allen, the resulting computer reconstructions of the gross morphology of sauropods were "wrapped tightly around the bones and therefore represent the minimum possible volume of the skeleton". This approach allowed the team to have a reproducible, unbiased protocol to reconstruct the appearance of the dinosaurs and several of their biophysical features.



Picture: K. Bates

Shorter necks, longer tails? Using a 3D computer-generated model of a sauropod, Bates and Allen tested different scenarios but always ended up with the same conclusion.

The analysis was applied to every sauropod, with a nearly complete skeleton, the group could scan, including some recently discovered near-complete Cretaceous titanosauriforms. Using their algorithm, the group obtained information about parameters, such as the total mass of the dinosaurs, their volume and the position in the body where most of the mass was concentrated, that is, the centre of mass. The data was normalised to allow comparisons among very different looking animals. The centre of mass of different segments of the body, for example, was divided by the estimated whole-body mass. Once normalised, the research team could see for the first time how size, shape and other biophysical parameters changed along the phylogenetic tree of sauropods.

Robust as a sauropod's front

Bates and Allen based their model of sauropod evolution on algorithmic reconstructions of digitised incomplete skeletons – but just how robust is it? Putting their model to the test, the team pushed their algorithms and assumptions to the limit. The convex hulls were widely expanded up to 21%; the body mass was computed for a wide range of possible scenarios, making the animals fatter or slimmer. For instance, in some cases they increased the mass of the trunk up to 50% or expanded the caudal or craniad segments of the body up to 100%. Pneumaticity, or the total empty space inside the bones, was identified for the first time as one of the variables that influences the sauropod body plan but, in general, no matter how many variations they applied, the phylogenetic model kept telling a consistent story.

In the middle of the Triassic, the centre of mass in many dinosaurs was moving closer to their hips, an adaptation for bipedalism. An unexpected turn of events occurred and any chance for bipedalism was reversed progressively during the middle Triassic, leading towards animals, with a centre of mass closer to their shoulders. This "craniad shift" in the late Triassic and early-middle Jurassic was reinforced in the late Jurassic. By that time, most sauropods were obligate quadrupedals. Needless to say, this process might have continued until the present but something like an asteroid decided to put a definite stop to dinosaur evolution in the late Jurassic. Interestingly, the moment when most sauropods became obligate quadrupedals, coincided with the appearance of the largest titans.

How to be a Jurassic titan

The team next looked closely at the anatomical changes that could produce the strong cranial shift of the centre of mass in sauropods. Neck enlargement, a key innovation in sauropod evolution, was found to be strongly correlated with gigantism. Hence, the scientists thought it would be interesting to test whether other animals with extremely long necks tend to have a craniad centre of mass. At first glance, modern giraffes look like good candidates, however, palaeobiologists maintain that most sauropods were not able to lift their necks upwards, like giraffes do. "The necks of some sauropods were so long that lifting them up would create an enormous pressure difference and the heart would have to do an enormous amount of work to pump blood up to the brain," explains Allen. Another feature of many sauropods, tail reduction, is also correlated with the centre of mass moving closer to the front. The tail in most reptiles is key to their locomotion because it contains the caudofemoralis longus (CFL) muscle. This muscle is anchored to the rear legs and is essential to retract the femur once a step has been taken, drawing the leg backwards. In crocodiles, it's responsible for the sinuous way of walking. In sauropods, however, the CFL-based locomotion seems to be less significant over time, as reduction of the size of the tail suggests.

Overall, Bates, Allen and colleagues show that gigantism in sauropods is correlated to a major craniad shift in their centre of mass in the late Jurassic. This probably was the result of anatomical changes, such as tail reduction and neck enlargement, conditions that could force them to adopt a form of quadrupedal locomotion based on their forelimb musculature and less on their tails.

Forward locomotion

Unfortunately for sauropod lovers, we're still far away from cloning dinosaurs and studying their biophysics *in vivo*. There are, however, other ways to test the theories that Bates and Allen put forward in the article. "Robotics is an excellent approach to understanding animal locomotion in general, not just locomotion in extinct animals like sauropods," explains Bates. "Building a sauropod-shaped, or a series of sauropod-shaped robots with slightly different morphologies, such as different neck lengths, might help us understand what effects differences in body shape had on the way sauropods moved," Bates concludes. "What I cannot create, I do not understand," Richard Feynman would have added.

In the history of palaeobiology, people went from digging up bones to reconstructing skeletons digitally by computer simulations. Maybe the next big step is indeed the creation of Saurobots. Will the dinos then rule the earth again?

Alejandrolvido

Photo: Pixabay/Pashminu

Publication Analysis 2007-2013

Nutrition Research

The Europe-wide cohort study, EPIC, exploring the link between nutrition and cancer, determines the "winners and losers" of the most-cited nations' and authors' ranking.

Burger with fries or ember bed-roasted veal wrapped in mushrooms with celeriac and apple, roasted celeriac jus and potato pithiviers (main dish at last year's Nobel Banquet)? What would you like to have now? Fact is, not everything we eat is good for us. Already, back in ancient Greece, Aristotle and Hippocrates noticed a link between diet and health/disease. "Persons in good health quickly lose their strength by taking purgative medicines, or using bad food," Hippocrates wrote more than 2,000 years ago. He also realised that "when more food than is proper has been taken, it occasions disease (…)".

The latter observation is today more topical than ever. Termed the global obesity epidemic or globesity, numbers of overweight or obese people have doubled throughout the last 30 years. According to the WHO, in 2014, almost two billion adults were overweight; of these, 600 million were obese. It's up to nutrition researchers to analyse these trends and associated health risks. But herein also lays nutrition researchers' big responsibility, as it is exactly those findings that shape governmental dietary guidelines and thus, have a direct impact on the public.

High and low impact

Now, without further ado, let's have a closer look at the scientific output of nutrition researchers in Europe between 2007 and 2013. For our nations' ranking, we, once again, relied on the specialist journals, as listed by the Web of Science database, under the subject category Nutrition & Dietetics. Interestingly, for a discipline with such a large impact, only a handful of specialist journals have an impact factor above 6. In contrast to the nations' ranking, the most-cited authors' ranking is, again, based on articles, reviews and proceedings papers published in all journals, multi- as well as monodisciplinary ones.

Sticking to food terminology, we will now serve the starters of this publication analysis, the nations' ranking. Famous for fish and chips, shepherd's pie and mint sauce, England is the unchallenged number one in European nutrition research. Then there are some surprises. Places two and three are occupied by Spanish and Italian researchers. Although this isn't so surprising these days. Among the European diets, the Mediterranean diet, with lots of olive oil, herbs and spices, has been shown to be associated with many health benefits and is thus at the centre of many public health studies. This study focus could also explain Greece's top position (12th).

Other nations can't quite keep up with that performance. Despite their world-famous "Schnitzel culture", Germany (5th) and Austria (19th) did not do very well in nutrition research. Sorted by citations per article, Scotland (24.4), The Netherlands (23.1) and Finland (22.9) are at the top. When compared to their US-American peers, European nutrition researchers again had a higher output and more citations but the citation-per-article ratio is, once more, in favour of US nutrition researchers.

After having digested the nations' performance starters, it's time for the main dish, the most-cited papers of nutrition researchers. Here, we decided to omit those papers that merely describe certain gene variants predisposing to diabetes or adiposity. The same limitation applies to the most-cited authors' ranking, in which scientists who mainly publish susceptibility studies have been excluded.

This puts two papers from the labs of Patrice Cani and Remy Burcelin in the top five (at positions 1 and 3). Both are about metabolic endotoxemia as a cause of obesity, diabetes and insulin resistance. The scientists noted, in mice, that a four-week high-fat diet chronically increased the plasma levels of lipopolysaccharide, an inflammatory reagent, from gut bacteria, two to three fold. "Metabolic endotoxemia dysregulates the inflammatory tone and triggers body weight gain and diabetes," Cani *et al.* discovered. Top papers on spots two and five also have something in common: both explore the relationship between adiposity/obesity and mortality. In Whitlock *et al.*, the researchers found that the median survival of persons with a body mass index (BMI) between 30-35 kg/m^2 (moderately obese) is reduced by two to four years; of persons with a BMI between 40-45 kg/m^2 (very severely obese) by eight to ten years. And last but not least, in 4th place, there's a single-author review about glucagon-like peptide 1 by its discoverer, Jens Juul Holst. This neuropeptide and gut hormone is released when we eat food; it stimulates insulin and inhibits glucagon secretion.

A true EPIC study

Ready for the delicious dessert? Panna cotta, crema catalana or Schwarzwälder Kirschtorte? European cuisine is versatile; the topics of the nutrition researchers in our top 30 most-cited authors' list are not. Almost all are epidemiology studies, trying to understand how certain foods and eating habits make us sick. A big portion of those publications present the results of the European Prospective Investigation into Cancer and Nutrition (EPIC) study. With more than half a million participants, it is one of the largest cohort studies in the world. Data collection began in 1993 in Spain, France, Italy and the United Kingdom; today, the EPIC biorepositories host more than nine million aliquots of plasma, serum, leukocytes and erythrocytes.

Intriguingly, not less than 15 most-cited nutrition researchers in our top 30 currently are Principal Investigators at local EPIC centres: Anne Tjønneland, 5th; Kim Overvad, 7th (Denmark); Françoise Clavel-Chapelon, 11th (France); Heiner Boeing, 4th; Rudolf Kaaks, 12th (Germany); Antonia Trichopoulou, 19th (Greece), Domenico Palli, 23rd; Salvatore Panico, 29th; Rosario Tumino, 20th (Italy); Göran Hallmans, 9th (Sweden); H. Bas Bueno-de-Mesquita, 13th; Petra Peeters, 15th (The Netherlands); Timothy Key, 22nd (Oxford) and our top duo Kay-Tee Khaw, 2nd and Nicholas Wareham, 1st (Norfolk). Add to this the study's coordinator, Elio Riboli (6th), and a few former (Jakob Linseisen, 26th) as well as deceased investigators (Sheila Bingham, 8th; Dimitrios Trichopoulos, 24th) and our top 30 list is almost complete.

Nutrition researchers number one and two, Nicholas Wareham and Kay-Tee Khaw, stand out not only with the high number of citations but also with the high number of publications – more than 500 for each one of them. This translates to publishing roughly two papers per week. In epidemiology, collaborative papers with hundreds of authors are rather the norm and not the exception. But can really everyone call him/herself an author of that study? "Not many of these individuals will have even seen the paper, and so the term 'author' seems meaningless and 'contributor' more appropriate," says COPE, the Committee on Publication Ethics. As Web of Science can't yet separate authors from contributors, we might take this ranking with a pinch of salt or rename it "most-cited authors/contributors in nutrition research".

Besides the epic EPIC study, more nutrition research has been going on in Europe. In France, Serge Hercberg (25th) and Pilar Galan (30th) worked on the SU.VI.MAX project, exploring the health effects of antioxidant vitamins and minerals, and the Nutrinet-Santé study: "a web-based prospective study on the relationship between nutrition and health and determinants of dietary patterns and nutritional status".

Also, the above-mentioned top paper authors, Jens Juul Holst (16th) and Patrice Cani (28th), made our top 30. Focussing on physiological aspects of food metabolism are Stephen O'Rahilly (17th) and Johan Auwerx (18th). The former is interested in elucidating the processes controlling energy intake and expenditure to get to grips with aetiology and pathophysiology of human metabolic and endocrine diseases. Auwerx wants to understand "how diet, exercise and hormones control metabolism". Last but not least, Michael Stumvoll (21st) focusses on genetics of adipositas, diabetes but also neuroendocrine control of appetite and the benefits of a Mediterranean diet on cognitive function in aging.

Personalised dietary advice

For now, nutrition research is predominated by large scale cohort studies, analysing effects of diet on health, on population level. The future of the discipline, however, might get a little more personal – personalised nutrition is the name of the game. Nutrigenetics and -genomics will help researchers decipher how one's own genetic makeup affects food metabolism and, *vice versa*, how nutrients affect gene expression. In a 2011 review, Australian nutrition researchers assert: "At the moment, there is a degree of public confusion and an immunity to messages that foster unpopular advice, such as 'get more exercise' or 'eat less calories'. Nevertheless, in the long term, these fields of endeavour may be the only way to optimise nutrition for optimal effects on health, wellness, and a slowing of the deterioration associated with the aging process" (*J Nutrigenet Nutrigenomics*, 4(2): 69–89).

Kathleen Gransalke

Europe...

Country	Citations	Articles	Cit./Art.
1. England	108,911	4,835	22.5
2. Spain	75,630	4,329	17.5
3. Italy	66,560	3,470	19.2
4. Netherlands	60,225	2,609	23.1
5. Germany	59,393	3,051	19.5
6. France	58,675	2,953	19.9
7. Sweden	32,761	1,454	22.5
8. Denmark	26,545	1,203	22.1
9. Switzerland	24,769	1,140	21.7
10. Belgium	24,303	1,100	22.1
11. Finland	20,504	896	22.9
12. Greece	19,239	957	20.1
13. Norway	18,251	922	19.8
14. Scotland	17,982	734	24.5
15. Ireland	14,822	753	19.7
16. Turkey	14,808	856	17.3
17. Poland	12,581	717	17.6
18. Portugal	11,700	608	19.2
19. Austria	9,384	480	19.6
20. Israel	8,662	429	20.2

Articles appearing between 2007 and 2013 in 'Nutrition & Dietetics' journals as listed by *SCImago* and Thomson Reuters' *Web of Science*. The citation numbers are accurate as of April 2016. A country's figures are derived from articles, where at least one author working in the respective European nation is included in the authors' list. Israel is included because it is a member of many European research organisations and programmes (EMBO, FP7 of the EU...).

... and the World

	Citations	Articles	Cit./Art.
Europe	500,794	27,317	18.3
USA	370,580	17,622	21.0
Canada	67,724	3,386	20.0
China	62,258	3,649	17.1
Australia	58,084	3,218	18.1
Japan	38,188	3,129	12.2
Brazil	28,015	2,724	10.3

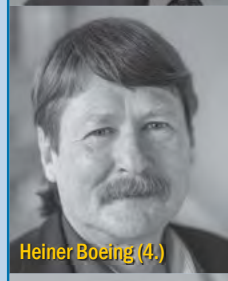
Publication Analysis 2007-2013 – **Nutrition Research**

Most Cited Authors...

		Citations	Articles
1.	**Nicholas J. Wareham,** Epidemiol, Univ Cambridge	37,064	569
2.	**Kay-Tee Khaw,** Publ Hlth, Univ Cambridge	27,018	540
3.	**Jaakko Tuomilehto,** Publ Hlth, Univ Helsinki (emerit.)	25,692	277
4.	**Heiner Boeing,** Epidemiol, German Inst Human Nutr Potsdam	15,189	433
5.	**Anne Tjønneland,** Danish Canc Soc, Copenhagen	14,726	491
6.	**Elio Riboli,** Publ Hlth, Imperial Coll London	14,610	368
7.	**Kim Overvad,** Publ Hlth, Univ Aarhus	14,340	469
8.	**Sheila Bingham,** Dunn Nutr Unit, Cambridge († 2009)	13,583	223
9.	**Göran Hallmans,** Biobank Res, Univ Umeå	12,419	306
10.	**Paolo Vineis,** Epidemiol & Publ Hlth, Imperial Coll London	12,366	300
11.	**Françoise Clavel-Chapelon,** Epidemiol & Populat Hlth, INSERM	12,095	306
12.	**Rudolf Kaaks,** German Canc Res Ctr, Heidelberg	11,851	345
13.	**H. Bas Bueno-de-Mesquita,** Natl Inst Publ Hlth & Environm, Utrecht	11,630	342
14.	**Carlo La Vecchia,** Epidemiol, Mario Negri Inst Pharmacol Res, Milan	11,482	395
15.	**Petra H. M. Peeters,** Publ Hlth, Imperial Coll London	11,049	334
16.	**Jens J. Holst,** Biomed Sci, Univ Copenhagen	10,753	327
17.	**Stephen O'Rahilly,** Clin Biochem, Univ Cambridge	10,529	124
18.	**Johan Auwerx,** Integrat & Syst Physiol, EPFL, Lausanne	10,513	121
19.	**Antonia Trichopoulou,** Hyg Epidemiol, Univ Athens	10,271	315
20.	**Rosario Tumino,** Canc Registry, Ragusa	10,220	349
21.	**Michael Stumvoll,** IFB Adipos Dis, Univ Leipzig	9,946	205
22.	**Timothy J. Key,** Epidemiol, Univ Oxford	9,941	240
23.	**Domenico Palli,** Canc Prevent & Res Inst ISPO, Florence	9,603	316
24.	**Dimitrios Trichopoulos,** Univ Athens († 2014)	9,495	216
25.	**Serge Hercberg,** Nutr Epidemiol, Univ Paris	9,338	204
26.	**Jakob Linseisen,** Epidemiol, Helmholtz Ctr Munich	8,514	190
27.	**Andrew R. Ness,** Oral & Dent Sci, Univ Bristol	8,360	119
28.	**Patrice D. Cani,** Louvain Drug Res Inst, Brussels	8,328	94
29.	**Salvatore Panico,** Clin & Expt Med, Univ Naples	8,140	204
30.	**Pilar Galan,** Nutr Epidemiol, INRA, Paris	8,106	134

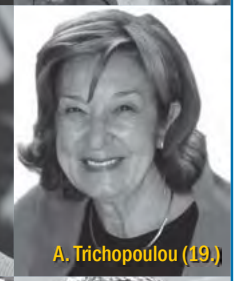

Nick Wareham (1.)


Kay-Tee Khaw (2.)


Heiner Boeing (4.)


Anne Tjønneland (5.)


Kim Overvad (7.)


A. Trichopoulou (19.)


Serge Hercberg (25.)
Pilar Galan (30.)


Patrice Cani (28.)

Citations of articles published between 2007 and 2013 were recorded up until April 2016 using the *Web of Science* database from Thomson Reuters. The "most-cited papers" had correspondence addresses in Europe or Israel.

... and Papers

	Citations
1. Cani, PD; Amar, J; Iglesias, MA; [...] Delzenne, NM; Alessi, MC; Burcelin, R Metabolic endotoxemia initiates obesity and insulin resistance. *DIABETES 56(7): 1761-1772 JUL 2007*	1,272
2. Whitlock, G; Lewington, S; Sherliker, P; [...] Qizilbash N; Collins R; Peto R Body-mass index and cause-specific mortality in 900 000 adults: collaborative analyses of 57 prospective studies. *LANCET 373(9669): 1083-1096 MAR-APR 2009*	1,235
3. Cani, PD; Bibiloni, R; Knauf, C; [...] Neyrinck, AM; Delzenne, NM; Burcelin Changes in gut microbiota control metabolic endotoxemia-induced inflamm. in high-fat diet-induced obesity and diabetes in mice. *DIABETES 57(6): 1470-1481 JUN 2008*	915
4. Holst, JJ The physiology of glucagon-like peptide 1. *PHYSIOLOGICAL REVIEWS 87(4): 1409-1439 OCT 2007*	911
5. Pischon, T; Boeing, H; Hoffmann, K; [...] Jenab, M; Ferrari, P; Riboli, E General and abdominal adiposity and risk of death in Europe. *NEW ENGLAND JOURNAL OF MEDICINE 359(20): 2105-2120 NOV 13 2008*	796

Antibody producer accepts civil penalty

Rigorous Punishment

What a twist! After years of wilfully ignoring the US Animal Welfare Act (AWA) [see *Lab Times* 2/2016, page 42, "The Dark Side of Research Antibodies"], Texas-headquartered antibody producer Santa Cruz Biotechnology (SCBT) has agreed to pay a fine of $3.5 million to the USDA (United States Department of Agriculture). In addition, its US dealer license for AWA-covered animal products will be terminated by December 31, 2016 as well as its registration as a research facility (in May 2016). This ends a smouldering fourteen-year conflict between the antibody supplier and the USDA.

Since 2002, USDA inspectors have repeatedly accused SCBT of numerous violations of the AWA, including the improper housing and insufficient medical care of thousands of rabbits and goats. Violations included a hidden, unapproved barn housing more than 800 additional goats and the disappearance, in autumn 2015, of all of the facility's rabbits and goats. These incidents resulted in three USDA complaints, which have now culminated in this rigorous, historic penalty.

SCBT will still be able to work with mice and rats – animals excluded from the AWA – and might therefore focus on monoclonal antibodies, which are still sold in Europe.

Sigrid März

Qiagen comes closer to Exiqon takeover

Tenacious Struggle

Qiagen's undertaking to acquire Danish Exiqon is close to the finshing line. Having already bagged 89 percent of Exiqon's share capital by June 2nd, there's just a hair's breadth keeping them from the magic 90 percent mark that gives takeover bidders the right to compulsorily gain the remaining 10 percent. Overall, the acquisition will cost Qiagen around €91 million. Exiqon's leadership, led by molecular biologist, Lars Kongsbak, supports the offer.

The Danish company, founded in 1995, sells tools for RNA analysis that complement Qiagen's RNA product line, according to Qiagen's CEO, Peer Schatz. Headquartered so far in Vedbaek near Copenhagen, Exiqon employs 100 people and generated sales of €22 million in 2015. After the completion of the acquisition, its public listing is to be settled.

Winfried Koeppelle

Thermo Fisher's next takeover

Closer to Proteins

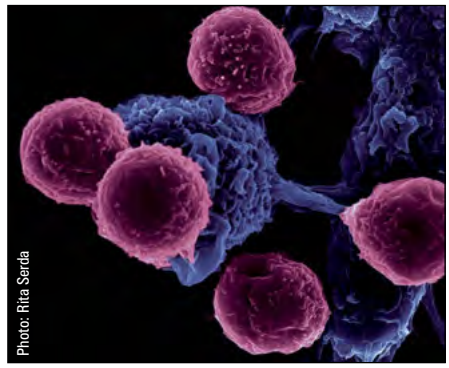


Photo: Rita Serda

Dendritic cells stimulated with adjuvant silicon microparticles interact with T cells, taken by a FEI-made Nova NanoSEM microscope (magnification 16,000-fold).

The list is almost endless: Affymetrix (DNA microarrays, California); Brahms (clinical diagnostics, Germany); Dionex (chromatography systems, California); Fermentas (restriction enzymes, Lithuania); Gibco (cell culture media, New York); Invitrogen (molecular biology reagents, California); Ion Torrent (DNA sequencers, Connecticut); Phadia (blood test systems, Sweden); Pierce Biotechnology (protein research products, Illinois); plus more than 200 additional companies that have been snapped up by Thermo Fisher in the last few years.

The US biotech provider, located in Waltham, Massachusetts, has grown to a labware giant that employs 51,000 people and sells products worth €15 billion per year.

Soon, Thermo Fisher will become even more powerful, if the planned acquisition of electron microscopy firm, FEI, goes smoothly. The €3.7 billion bid is to, "complement Thermo Fisher's existing mass spectrometry business, particularly in its work on structural proteins and biopharmaceuticals", according to Thermo Fisher's CEO Marc Casper. Combining mass spectrometry data with the structural information provided by cryo-EM provides a picture of both the shape of a protein as well as an understanding of the arrangement and orientation of individual proteins within a whole structure.

Hillsboro, Oregon-headquartered FEI has over 3,000 employees. The takeover is expected to be completed by early 2017.

Winfried Koeppelle

Scottish University creates biotech firm

Growing Insulin Islets

Patients with type 1 diabetes should take a good look at northeastern Scotland. To improve the treatment of this metabolic disorder, caused by an autoimmune attack that leads to the loss of insulin-producing β-cells, the University of Aberdeen recently announced the creation of a start-up.

The newborn Islexa is dedicated to producing laboratory grown pancreatic islets, the organoid structure responsible for insulin production. Islexa's scientists seek to reprogramme donated pancreatic tissue into fully functional islets. So, type 1 diabetics would no longer need to administer insulin. According to endocrinologist Kevin Docherty, who is one of the participating key scientists, the technology is based on converting pancreatic tissue into functional islets. Thus, no stem cells are needed.


Photo: Donald Thomas

The Institute of Medical Sciences at University of Aberdeen, Scotland.

If it works in the way the Scots envision, a healing islet transplantation could become reality for thousands with type 1 diabetes. Currently, only 50 British diabetics per year, at the very most, are able to receive an islet transplant. This is due to medical/technical problems involved in extracting the islets, as well as to a shortage of suitable donor organs.

Winfried Koeppelle

Life sciences fund obtains €245 million

Focus on Europe

LSP's founder, Martijn Kleijwegt

Photo: LSP

Venture capital firm, Life Sciences Partners (LSP), based in Amsterdam with offices in Munich and Boston, has closed its new life sciences fund, LSP 5. With a respectable €245 million raised, LSP 5 is one of the largest life sciences funds in Europe.

What is the money intended for? According to LSP, they are primarily focussed on European biotech and looking for "around 20 companies across drug development, medical devices and diagnostics".

The fund has already invested in nine companies, such as Irish medical device company Neuravi (working on acute ischaemic stroke), Swiss-based immuno-oncology company Nouscom and Kuros Biosciences, focussed on tissue repair and regeneration.

LSP has raised more than €1 billion since its foundation in 1998, feeding a lot of (now) more or less well-known biotech companies with venture capital. These include: Crucell, Merus and Sapiens (all from The Netherlands); Argenx and Movetis (Belgium); Activaero, Jerini and U3 Pharma (Germany); Okairos (Switzerland) and Zealand Pharma from Denmark. In addition, LSP was involved in taking Qiagen public in 1996, selling Kudos Pharmaceutical to AstraZeneca in 2006 and performing a €90 million IPO with Prosensa in 2013.

René Kuijten, a co-owner of LSP, commented that the appetite for investment in life sciences in Europe has increased significantly over the years. He added that, "unlike the IPO market, which has been tough for biotechs in 2016, the VC environment is in rude health." Winfried Koeppelle

Belgium's Argenx scores again

Bright Future

As always, the Belgian national football team is joint favourite at the 15th European Football Championship. And as always, *De Rode Duivels* will have little chance against great nations like France, Germany and Spain. But at least in the biotech field, Belgium is more successful. Take, for example, Argenx's last financing round. The Ghent-based antibody engineering company recently raised €30 million, just a few weeks after launching its smash hit, a huge preclinical immuno-oncology deal with US giant Abbvie, worth €600 million.

So, it should now be dead easy to complete preclinical testing of Argenx's immuno-oncology candidate, ARGX-115, and, if all goes well, then transfer clinical testing to Abbvie. ARGX-115 is intended to treat all kinds of cancers as it stimulates a patient's immune system after a tumour has suppressed it. -wk-

Bayer's 56 billion euro offer for Monsanto

Suboptimal Publicity

Pesticides like Roundup haven't such a good reputation as Bobby Cars.

Photo: Erntteservice Schneeweiß

Photo: Simba-Dickie

There really are more enjoyable takeover targets – companies with a positive, generally more accepted image. Let's say Lego. Or the Simba-Dickie Group, which produces the most environmentally friendly car in the world, the Bobby Car.

But Bayer, the German chemical and pharmaceutical group, focussed on, of all things, Monsanto, making a €56 billion takeover offer for the US agricultural company on 10th May.

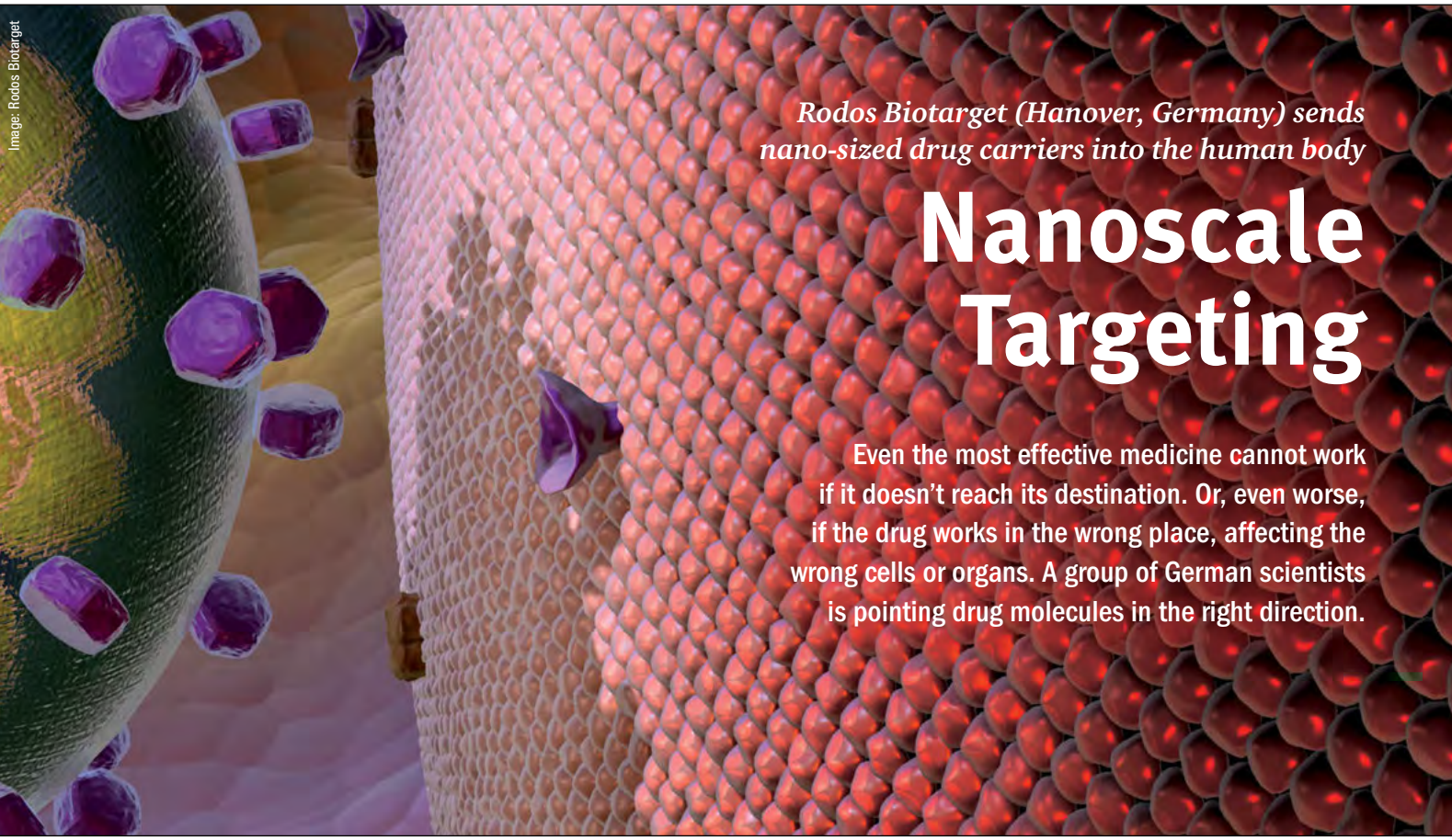
You must remember Monsanto, the ill-famed seed company, which sells the controversial broad-spectrum herbicide, glyphosate (Brand name: Roundup). With glyphosate alone, being one of the most-used agricultural herbicides, Monsanto makes $2 billion per year – and, in the process, has become the ill-reputed target of dozens of environmentalists and non-government organisations (NGOs).

Monsanto has done everything possible in the last 80 years to build up a "bad guy" image. The company assisted in the development of the first nuclear weapons in the 1940s, began manufacturing DDT in 1944, turned into the world's most important producer of polychlorinated biphenyls (PCBs) by 1977, and was, together with Dow Chemical, the main producer of the herbicide Agent Orange, which during the Vietnam War was used to destroy the enemy's crops and vegetation in "herbicidal warfare". In addition, Monsanto has executed field trials of genetically modified crops from 1987 onwards and is accused by Greenpeace and other NGOs of the, "aggressive acquisition of patents, control of farmers, and accepting the contamination of large areas with GM crops".

By the way, the WHO classified glyphosate as "probably carcinogenic in humans" in March 2015, while the European Food Safety Authority concluded in November 2015 that, "the substance is unlikely to be genotoxic or to pose a carcinogenic threat to humans." Oops! A fierce glyphosate dispute is now taking place around the issue of whether to allow or ban its use on European grain fields. No wonder that this controversy overshadows Bayer's billion euro offer to buy Monsanto.

It was certainly not the smartest move on the part of Bayer's brand new head, Werner Baumann, to startle the German public with the takeover offer, just ten days after he had taken over the baton from former Bayer CEO, Marijn Dekkers. But he had no choice. Monsanto's executives had made him an offer just a few days before in exactly the opposite direction, by seeking to acquire Bayer's agricultural division, Bayer Cropscience. Baumann turned the tables and so, the hunted became the hunter.

Nevertheless, most industry experts think that €56 billion is too much and will financially weaken Bayer. In addition, the Germans probably have to restructure Monsanto after a successful acquisition. Together with the public headwind and other parties that are interested in Monsanto, such as German BASF, Baumann has to hurry up with the merger procedure – or should stop it. If successful, however, he has to transform the, "world's most hated company" into a publicly accepted and still lucrative subsidiary. There are easier tasks. -wk-



Rodos Biotarget (Hanover, Germany) sends nano-sized drug carriers into the human body

Nanoscale Targeting

Even the most effective medicine cannot work if it doesn't reach its destination. Or, even worse, if the drug works in the wrong place, affecting the wrong cells or organs. A group of German scientists is pointing drug molecules in the right direction.

Early summer 2012, when Robert Furch was facing his young company's doom, was a truly awful time. With venture capital running out and no investor in sight things looked bad. Most start up entrepreneurs can sympathise with this nightmare. But in the end, the situation wasn't as bad as it seemed. Furch and his colleagues on the board dug deep into their own pockets to bridge the finance gap for a few months. Good decision! Before the summer of 2012 ended, by 1st August in fact, they had secured a comforting, external follow-up financing. Since then, their drug delivery company, Rodos Biotarget, has been back on track.

Furch, a 47-year-old biochemist who trained mainly at the University of Heidelberg, Germany, and later abroad in Spain, USA and Brazil, doesn't give the impression of somebody prone to dramatic exaggeration. Your *Lab Times* reporter had to inquire several times before hearing about the funding shortfall described above. The history of Rodos Biotarget, however, began earlier, of course. Much earlier, in the late 1980s in Southern California.

Michael Scolaro, a neuropsychiatrist from Los Angeles, was one of the first to specialise in the treatment of patients with AIDS, after the immune deficiency syndrome was first clinically observed in LA in 1980 and then began its global spread (on the discovery of AIDS: Michael Gottlieb, *Morbidity and Mortality Weekly Report*, June 5, 1981; Gottlieb *et al*., *N. Engl. J. Med*. 305 (24): 1425–31).

In the eye of the HIV storm

Scolaro soon became a key clinician in the treatment of AIDS in the *City of Angels* area. He founded a virus research institute there that was later transformed into the non-profit "Let There Be Hope Medical Research Foundation" (aka "LTBH institute"). Scolaro's objective was to target antigen presenting cells of the immune system that act as hidden reservoirs for viral DNA.

In the late 1980s, for example, he investigated whether the chemotherapeuticum, Daunorubicin, is effective in treating Kaposi's sarcoma in AIDS patients when it is packaged in liposomes. He also collaborated with a group of scientists from Göttingen, Germany, to inhibit HIV viral replication using the immunoliposomal targeting of infected CD4 cells. Robert Gieselervon der Crone and Jörg Ruppert, two young German biologists who both took their PhD at the University of Göttingen in 1991, were part of the LA team.

Nowadays other methods (i.e. polymeric micelles) exist that can transport a drug to its target in the body. But liposomes have nevertheless emerged as the most appropriate and thus most commonly used vehicles. They are biocompatible, non-toxic and non-immunogenic. The only problem is their prompt uptake and clearance by the endogenous reticuloendothelial system (RES) *in vivo*, and their relatively low stability *in vitro*. To prevent this, the half-life of liposomes is often elongated by adding polyethylene glycol (PEG) to their surface.

Improving liposomal carriers

Back to LA, where, in the late 1990s, Scolaro's group became more and more experienced in the targeted liposomal ▶▶

Freelance Biobusiness Writers Wanted

(Applications from the UK, France & Scandinavia especially welcome)

Please contact:
wk@lab-times.org

Rodos Biotarget's founders, Marcus Furch (left) and Robert Gieseler-von der Crone, are doing their very best to commercialise their nanocarrier platform technology.

Photo: Nishika Sahni

delivery of therapeutics. They developed tricky nanocarriers, labeled with carbohydrate molecules on their surface and similar to the viral envelope carbohydrates of pathogens which are used to attach to key immune reservoir cells. After mimicking the virus' means of entry, the liposomal nanocarriers should release their payload: the therapeutic agent.

To put it briefly, it worked. Gieseler-von der Crone, then working as a chief scientist at Scolaro's LTBH Medical Research Institute in Beverly Hills, together with Scolaro himself and others, finally developed a "well-tolerated and poorly immunogenic" transfer vehicle, the so-called CLR-Targosphere. It's a sophisticated invention. The nanoparticle's core is aqueous (lipophobic), but it is wrapped in a lipid bilayer capsule, embedded with targeting anchors that enable the carrier to bind solely to C-type lectin receptors (CLRs).

The ideal target

Or, more simply: a tiny parcel with an accurate address label, ready to be filled with drugs and sent to the sick organ.

But why, of all things, did they address C-type lectin receptors? Well, those receptors are the ideal target in most diseases. They are expressed on the surface of antigen-presenting cells (APCs), which are critical to specifically recognising pathogens, distinguishing between healthy and malignant cells, and maintaining immunotolerance. Thus, if it were possible to deliver active agents selectively and efficiently to APCs, the immune system of a, say, cancer patient could be mobilised against the malignant disease.

Scolaro's Targosphere technology seemed to offer huge potential. Ten years ago, any commercial clinical application was still a daring dream for the future. Scolaro wisely filed the relevant patent applications in 2004/2005 to protect intellectual property concerning the site-specific intracellular targeting of key immune cells.

And now the story is shifting to the opposite side of the globe: to Germany.

2008: Two companies are founded

In 2007, the time had come to commercialise the Targosphere technology. Gieseler-von der Crone, Ruppert and Scolaro participated in the Science4life Venture Cup, a German businessplan contest for scientists who intend to found a biotech company. They didn't win but placed as a respectable fifth, with their nanocarrier platform technology intended to treat infection (HIV/AIDS, hepatitis C, tuberculosis and others), cancer and autoimmune diseases. And they met a young German biochemist at the Science4life events, acting as jury member and coach, who had already gained some experience in the pharmaceutical industry.

His name was Marcus Furch.

One year later, Gieseler-von der Crone, Ruppert, Scolaro and Furch were ready to found a startup biotechnology company, Rodos Biotarget, in Hanover. Scolaro's LTBH Foundation sold the intellectual property to the young German startup, and in North Myrtle Beach, South Carolina, a company with similar objectives was formed: Augustus Biotarget. Both companies cooperate closely but are independent from each other.

When it comes to establishing a biotech company, what is at least as important as sophisticated technology? Without question – money! So far, Rodos Biotarget has raised about €6-7 million via conventional investment rounds, government grants, private VC investors and individual business angels.

To increase this sum, however, Furch and Gieseler-von der Crone broke new ground: they used crowdfunding.

Seeking a crowd

Kraut what?! Well, crowdfunding, a form of alternative fundraising which sounds progressive but in fact has a long history. Take French philosopher, Auguste Comte (1798-1857), who once requested financial support from fellow citizens for his activities as a philosopher. Or the monumental base for the Statue of Liberty, that could only be built after a campaign successfully attracted small sums from about 160,000 donors.

Today, crowdfunding is mostly performed via internet-mediated platforms, of course. Nevertheless, it's an unconventional approach, especially for biotech companies. But there are successful forerunners, such as Dresden, Germany-based Riboxx Pharmaceuticals. Between July 2014 and January 2015, Riboxx raised €1 million from 928 single investors for the first clinical test of an immunodrug to prevent cancer recurrence.

Furch would be very pleased if it works with Rodos Biotarget in a similar way. He and Gieseler-von der Crone are seeking around €1 million, too, and like Riboxx, they will post their offer on the *Seedmatch.de* platform.

"Around 1,000 or 1,500 euros each"

"People can invest from at least 250 to a maximum of 10,000 euros per individual", Furch says, "but the typical crowdfunding investor gives around 1,000 or 1,500 euros".

That means that his company has to inspire 300 to 400 private individuals with their campaign to achieve the target of a million. In an effort to persuade investors, Rodos Biotarget has already produced animated films that explain their nanocarrier technology. Additionally, there are giveaways waiting for each crowd investor: exercise balls in different sizes, depending on the amount that the investor has given, symbolising Targosphere bullets.

"It's important to explain our sophisticated technology, especially to the 'common' crowd funder who usually isn't skilled in molecular biology or other natural sciences", explains Furch. "We won't get money by those people if they don't understand the relevance of what we are doing. So we have to simplify, to visualise."

Furch is aware that such 'educational work' might be laborious, but it can also bring unexpected advantages, "We are forced to improve our external communication. To explain complicated contexts, you must hundred percent understand what you are doing. And this will make it easier, as a welcome side effect, to communicate with 'big' investors in conventional financing rounds."

The time frame is also shorter when it comes to crowdfunding. Conventional venture capital is usually intended to bridge a two- or three-year period, and thus the results may require the same time. Crowd investors, however, are less patient, Furch believes. So he and Gieseler-von der Crone had to come up with a research programme that can be achieved in just 12 months.

Clinical trial soon to be started

Finally, the Germans designed a brief low-risk clinical phase I study that should be completed within a year. Its primary goal is to demonstrate that Rodos' nanocarriers are harmless and induce no, or at least low, side-effects. In addition, the kinetics and metabolics will be analysed.

The trial will be undertaken in about 20 healthy testers and contain three arms: the first group will be administered unfilled Targospheres, the second group gets Targospheres filled with antibiotics, and the third group will take a placebo.

The trial will take place in the UK and has already been approved by the British regulatory authorities (MHRA). It is due to start as soon as the fundraising is completed.

The Germans aren't betting on crowdfunding alone, of course. In parallel, they are preparing their next financing round, aiming to raise up to €15 million. With this fresh money, they hope to reach clinical stage II.

Waiting for the big deal

As we have heard, Targospheres are well-tolerated and poorly immunogenic. Moreover, they cross the blood-brain barrier and can be potentially used to deliver neurodrugs. They can already be produced in defined sizes and offer an excellent payload capacity, says Furch. They have been fully preclinically validated and, at least in animals, no adverse effects on major organ systems have been observed. And, they can be produced in huge quantities. Furch mentions reassuringly that the production of the CLR-Targospheres has been scaled up to industrial batch sizes of greater than 20 litres under cGMP conditions.

Sounds great, but where is the applause? Are no drug developers interested in such a formidable carrier system?

Some are already on board, assures Furch. Since the end of 2014, for example, a pharmaceutical company from abroad – its name is confidential, he regrets – has been cooperating with Rodos Biotarget, aiming to make common antibiotics more effective. By using the German nanocarriers, they hope to address the inside of those cells where pathogens hide – and turn allegedly 'ineffective' drugs back into functioning ones.

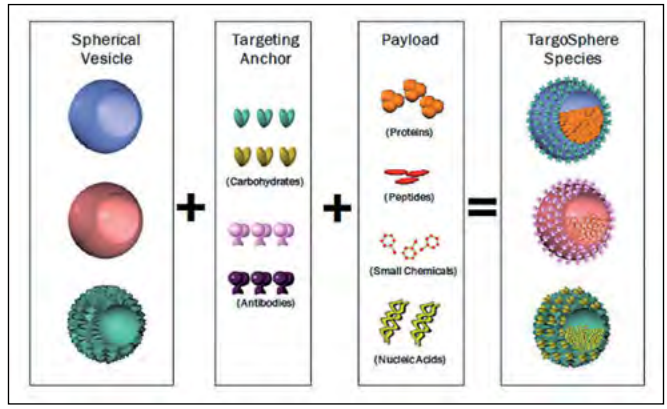


Image: Rodos BT

The Targosphere carriers can be configurated in different ways, to enable multiple applications and therapy options.

Turning the ineffective into functioning

In a few years, there may be a scrum of anxious prospective customers ringing the bell at 23 Feodor-Lynen-Street, Hanover Medical Park. After all, the pharmaceutical industry urgently needs appropriate cargo ships to transport their superb novel drug compounds into the human body, to the right organs and into the right cells. Rodos Biotarget might be able to help.

Winfried Koeppelle

Freelance Biobusiness Writers Wanted

(Applications from the UK, France & Scandinavia especially welcome)

Please contact:
wk@lab-times.org

Theranos founder, Elizabeth Holmes

Biotech "self-made woman" dragged down by dubious affair

Climb Fast, Fall Hard

By touting their "revolutionary" blood tests, diagnostic service provider Theranos became the hottest US biotech venture, with the company valued at €8 billion. There's growing suspicion, however, that their "Edison" technology, named after America's most famous inventor, is a dubious house of cards.

When it came to US healthcare company Theranos and its high-flying 32-year-old founder, Elizabeth Holmes, no superlative seemed sufficient to describe her tremendous genius. Holmes is, "shaking up the laboratory testing market" (*San Francisco Business Times*). The, "truly exceptional inventor fundamentally revolutionises blood testing and analyses" with her, "ground-breaking ideas" (Benoît Battistelli, President of the European Patent Office), making the 32-year-old a "power woman" and "the world's youngest self-made billionaire" (*Forbes*).

Wow! More about this marvellous woman, please.

Holmes founded a biotech company, Theranos, based in Palo Alto, California, 13 years ago, in order to place her "medical sensation" (The European Patent Office) on the market. Despite being a Stanford University dropout with no expertise in anything, she developed a tricky microfluidics blood-testing device for early illness detection as well as for disease prevention. Brilliant. According to Holmes, this device just requires a few microlitres of blood obtained via harmless fingerprick, rather than using a huge, 30-millilitre-vial and common, uncomfortable venipuncture.

Holmes named her ingenious blood-testing device Edison. Yes, after Thomas Alva Edison, the famous inventor of the phonograph and the long-lasting electric light bulb. As the following shows, it is possible that Holmes' fame will be significantly shorter than Edison's.

Illustrious investors

Patients certainly prefer not to be panicked by frightening needles. A minimised and automated technology, such as that promoted by Theranos, could also make routine diagnoses faster and cheaper. There are about 200 different laboratory tests on their list, ranging from routine blood analyses to advanced genetic tests. And the prices are certainly attractive – just a few dollars for an analysis that would previously have cost up to $800.

But the Edison device won't halt climate change. Theranos also hasn't found a miracle cure for AIDS, Alzheimer's disease and cancer. No, they just offer a simple new tool for blood diagnosis – somewhat better than most ordinary devices but certainly no killer application to revolutionise the entire medical industry.

For this reason, it's more than surprising that such an ordinary product has raised over $400 million (other sources even state up to $750 million) from US investors, pushing the company's value up to €8 billion within a few years. Illustrious individuals provided Theranos with venture capital, including, for example, Mexican business magnate Carlos Slim, one of the richest people in the world, as well as Oracle founder Larry Ellison, giving Holmes the capital needed to grow a huge company with a staff of 700 and a 25,000 square metre facility in Newark, California. The company's board of advisers is also crowded with impressive names, listing no fewer than six US elder statesmen.

It is strange, then, that this ingenious junior inventor has had no professional education (she left University at the age of 19 without a degree), and that she has never published any technical details about her invention. Somehow, Elizabeth Holmes and her Edison thing were a dubious black box from the very beginning.

Publicly pushed blind enthusiasm

Most Europeans don't appreciate such blind enthusiasm. In inspirable America, however, the Theranos hype turned Holmes, who owns 50 percent of the company, into a multi-billionaire. Or more precisely, into the youngest female billionaire in the world. Again and again, Holmes was praised by enthusiastic cover stories in leading US magazines such as *Forbes*, *Time* and *Fortune* (see above). Holmes made it into the "TIME 100 Most Influential People in the World in 2015", she became one of eight "Women of the Year" in *Glamour* magazine, and even the US president himself, Barack Obama, appointed the high-flying wonder woman as a "United States ambassador for global entrepreneurship".

But that's America: with enough public hype, gray mice are puffed up into magnificent unicorns. And there really was a lot of hype surrounding this awesome young woman from Washington, D.C., who, "invented a way to run 30 lab tests on only one drop of blood" (*Wired* magazine) and was going to be "the next Steve Jobs or Bill Gates" (according to former US Secretary of State, George Shultz).

Is this just America?

Holmes' fame finally reached Europe. She was named as a European Inventor Award 2015 finalist by the European Patent Office (EPO), in the somewhat strange category of "Non-European Countries". EPO President Benoît Battistelli himself praised her as "a truly exceptional inventor", and Holmes' "revolution based on a drop of blood" as having "the potential to fundamentally change the healthcare market" and, "move the world forward".

In the run-up to the award ceremony on 11th June 2015, the EPO also published a promotional video about Theranos, in which the unskilled dropout Holmes was

described as a "microfluidic specialist" and as a "US biotech genius", about whom the public would hear much more in the future.

In the end, Holmes didn't win the European Inventor Award 2015. But in a way, Battistelli was right: the public would soon hear much more of Holmes.

October 2015: a radically revised view

Interestingly enough, it was one of her greatest admirers that started challenging this perception of the alleged miracle company. In October 2015, *The Wall Street Journal* (*WSJ*) questioned the reliability of Theranos' technology, backed by several medical experts (bearing in mind that, two years ago, the same *WSJ* hyped, "the breakthrough of instant diagnosis", speculating that Holmes' inventions could "upend the industry of laboratory testing and might change the way we detect and treat disease"; *WSJ*, 8th Sept 2013).

Also in October last year, the Food and Drug Administration (FDA) conducted a surprise inspection and concluded that major sections of Theranos' test equipment weren't authorised for the relevant applications. Only one test system and test for the herpes simplex 1 virus IgG was regulatory approved, according to the FDA, while all other offered tests (about 200 in all) were neither tested for accuracy nor approved and thus used without the FDA's authorisation. Now, the FDA is demanding that all Theranos' technology goes through the regulatory process before proceeding with their testing.

Using competitor technology…

Holmes refutes all allegations. She maintains that Theranos had publicly reviewed the test results (and thus proven their accuracy) – an obviously groundless claim, given the fact that Theranos didn't ever release data showing reliability and accuracy. No one from outside the company has ever verified Theranos' technology – or, better put, Holmes has so far omitted to provide any external review. Apparently there was no independent auditing, no benchmark testing, no scientific publication in a peer reviewed journal – but a lot of interviews and gossip in *Vanity Fair* and *Glamour* magazine.

Even worse, Holmes finally had to admit that the vast majority of Theranos' tests hadn't been executed by their superb Edison device but by a foreign machine. No joke! Theranos had secretly filled up the alleged blood "microsamples" with water and then analysed them with common medical diagnosis machines, developed by German competitor, Siemens.

On 18th May, the *WSJ* reported that Theranos has even voided all the blood tests carried out by Edison devices in 2014 and 2015. It could "not be ruled out that doctors have taken incorrect treatment decisions, based on the initial results".

To add insult to injury, two alleged partnerships with GlaxoSmithKline and Pfizer turned out to be wishful thinking. Both companies affirmed that they didn't cooperate with Theranos – in contrary to what Holmes had claimed.

Taking all of this into account, the company's central claim of being able to detect and measure hundreds of therapeutically relevant compounds hidden in a few drops of human blood, is being called into question. Everything points to the idea that Holmes' hyped finger-prick technology is a waste of effort – because it might be less powerful than the common, already available diagnostic technology.

The Theranos board of advisers reads like a Who's Who of US politics and the military, featuring, among others (from left), William Perry (88, Secretary of Defense under Bill Clinton), George Shultz (95, former Secretary of State) and Henry Kissinger (93, Nobel Peace Prize laureate and former Secretary of State).

A €4 billion fortune drops to zero

In 2015, *Forbes* magazine had estimated Theranos' worth at $9 billion, making Holmes one of "America's Richest Self-Made Women" because of her 50 percent stake in the company. On June 1st, Holmes' virtual fortune of $4.7 billion dropped to zero within a mouseclick after the magazine's editors revised their estimate of Theranos' worth to only $800 million due to the bad news. If the struggling company were liquidated, Holmes shares would be worth nearly nothing, the editors argued.

The FDA is pressing hard on Holmes, too, by seeking to revoke Theranos' licence, banning her for two years from owning or operating a blood lab. And both the federal prosecutors as well as the Securities and Exchange Commission are running criminal investigations into Holmes and her company.

So it looks as if the whole Theranos hype will soon culminate in a record-breaking write-off – leaving behind displeased investors, excessive litigation and a disenchanted Elizabeth Holmes (perhaps behind bars, as some insiders predict).

The enigmatic Edison device

It's more than curious that no one questioned for such a long time the secretiveness around the Edison blood testing device. So much excitement, so much promise, but nothing known about the procedure or technology with which the company was working. A few droplets of blood are transferred into a mystical black box, just wait a little moment – et voilà, the result, much faster and far cheaper than anything else. Seriously, who would believe this crap?

Obviously, many influential rich people believed exactly this crap. Holmes' absurd assertions proved the old wisdom that the more incredible your claim, the more likely it is that many will believe you.

The fact that Holmes is blonde, pretty and female might have helped. Just take a look at the Theranos board of advisers. It reads like a Who's Who of US politics and the military, featuring Henry Kissinger (93, Nobel Peace Prize laureate and former Secretary of State), William Frist (64, former US Senate Majority Leader), George Shultz (95, former Secretary of State), William Perry (88, Secretary of Defense under Bill Clinton), Gary Roughead (65, retired US Navy admiral) and Samuel Nunn (77, Senator for Georgia for 24 years).

No idea of blood diagnosis

The problem is that those distinguished men know quite a lot about politics and warfare but nothing about blood diagnosis.

Holmes also meets the criteria of dropping out of college and founding a global player – just like Bill Gates, Steve Jobs and Mark Zuckerberg – as well as being omnipresent in the media (but never saying anything of scientific substance!). Last but not least, she is living an adequate lifestyle (she is vegan).

Someone like her must be ingenious, eh?

Winfried Koeppelle

Product survey: Thermocyclers

Alternative Heat Production

What have hot waterbaths, Peltier elements, hot air, electric resistance, magnetic induction, Ranque-Hilsch vortex tubes, infrared light, lasers and photons in common? They may all be used as heat sources in thermocyclers.

It's been three years since our last product survey on thermocyclers (see *Lab Times* 4/2013, p. 47), so what has happened since in the world of PCR machines? Well, it is still dominated by Peltier heated block cyclers but alternative heating concepts are gradually gaining ground. And there is another trend that cannot be overlooked: the number of small, easy-to-use, portable, pocket-sized thermocyclers suitable for field studies or point-of-care diagnostics is growing rapidly.

Garden variety block cyclers are equipped with Peltier heated aluminium or silver blocks, delivering heating rates of about 2 to 5°C per second and slightly lower cooling rates. Coating the silver block with a thin gold layer considerably enhances ramp rates, especially in combination with thin wall propylene tubes, which mould into the block holes like a second skin. High end block cyclers based on this concept, such as Analytik Jena's SpeedCycler^2, may obtain ramp rates of 15°C/s, which are amongst the fastest of current Peltier heated block cyclers.

But even the most sophisticated heat transfer techniques cannot hide a major drawback, which has haunted block cyclers since their inception in the late nineteen-eighties: due to material inconsistencies and edge effects, the temperature is not evenly distributed throughout the heating block. Typical deviations from perfect block uniformity range from 0.3 to 0.5°C at PCR-relevant temperatures of 72° and 95°C. That may not sound too bad but in combination with other common inaccuracies of block cyclers, such as over- and undershooting temperatures, even slight variations in block uniformity may contribute to significant errors in PCR experiments.

Block uniformity has been considerably improved by a technique originally developed in Axel Scherer's lab at the California Institute of Technology in 2006. His group came up with the idea to hollow out the silver block and fill the cavern with a thermally conductive liquid that is pumped through the hollow block during PCR cycling. The rapidly circulating fluid transfers the heat generated by the Peltier elements evenly across the block, providing a block uniformity of 0.1°C.

Hollow block cycler odyssey

The hollow block cycler has gone through quite an odyssey since Scherer's first attempts to commercialise it via the start-up company Helios, in 2007. Helios was acquired by NGS giant Illumina in 2010, who initially sold the hollow cycler as the Eco Real Time PCR system but quickly lost interest in the thermocycler market and discontinued the Eco cycler in 2013. The NGS company sold the intellectual property rights for the Eco cycler to the British PCR distributer, PCRmax, who was purchased by the life science supplier, Bibby Scientific, in 2014. Shortly after the acquisition, Bibby Scientific relaunched the hollow cycler under the brand names Eco 48 (PCRmax) and Prime Pro 48 (Techne).

Scherer's concept to enhance temperature uniformity with a fluid filled heating block is pretty smart. The easiest way to eliminate erratic temperature deviations in thermocyclers is, however, to completely abandon Peltier heated aluminium or silver blocks and switch to alternative heating techniques. One of the first implementations of this idea was an air-heated carrousel qPCR cycler, introduced by the small US biotechnology company Idaho Technologies in 1997, under the brand name LightCycler. The heating concept of the LightCycler is both simple and elegant: the qPCR reactions are carried out in thin, glass capillaries inserted into the holes of a rotor, spinning in an air-heated chamber. A small fan at the bottom of the chamber draws ambient air into the compartment that passes an electronically-regulated heating coil, placed in the inlet channel on top of the chamber. After circulating around the glass capillaries, the air leaves the chamber via laterally oriented outlet channels. To ensure fast cooling and heating rates, the fan rotates at lower speeds at the heating phases of the PCR cycles and accelerates during cooling. Detection of the amplicons is achieved by a laser beam focussed on the tip of the respective glass capillary in the measuring cell. To this end, a stepper-motor gradually rotates the carrousel to put the capillaries stepwise in the optical axis of the laser beam.

Rotary cycler with a twist

The then revolutionary heating concept of the LightCycler instrument quickly sparked the interest of Boehringer Mannheim. The German pharmaceutical corporation acquired the property rights for the technology, shortly before it was taken over by Swiss pharmaceutical giant Hoffmann-LaRoche, now Roche, in 1997. A few years later, John Corbett Senior, an engineer and inventor from Down Under, presented a similar rotary cycler system – but with a few little twists. Corbett's rotary cycler, called Rotor-Gene, is based on a shallow centrifuge rotor, perforated like Emmental cheese, to allow maximal air circulation. The PCR is accomplished in standard, thin wall tubes looming out of the rotor places into an air-heated chamber. Heating and cooling of the chamber is achieved similar to the LightCycler by ambient air, forced into the compartment by a fan. But the detection of the PCR products is done slightly different. PCR tubes spin continuously and pass the excitation optics every 150 milliseconds to deliver a fluorescence signal that is detected by a photomultiplier tube. Rotary cyclers offer very fast ramp rates of up to 20°C/s but what makes them really stand out are untouched temperature uniformities of less than 0.02°C, e.g. for the Rotor-Gene.

Similar to the managers of Idaho Technology, John Corbett Sr converted his air-heated rotary cycler into money and sold his company Corbett Life Sciences together with the Rotor-Gene to Qiagen in 2008. But Corbett obviously didn't want to rest on the 70 million dollars that he cashed in with the Qiagen deal. Together with his son, John Corbett Junior, he launched a new qPCR

thermocycler, based on magnet induction heating, at the Biotechnica trade show in October 2015. As Corbett Sr told a television team filming his new Magnetic Induction Cycler (MIC) at the booth of his newly established company Bio Molecular Systems, he got the idea for his invention while heating coffee water on a magnetic induction cooktop. The underlying concept of the MI-Cycler is very straightforward: The Corbetts' still rely on the tried and true rotary system of the Rotor-Gene and the fan for cooling but have substituted the heating coil with an electromagnetic coil that surrounds the aluminium rotor of the MIC. An alternating current flowing through the coil induces an alternating magnetic field around the coil that pervades the electrically conducting aluminium rotor and induces Eddy currents inside the metal. Due to the electric resistance of the metal, the induced Eddy currents instantly heat up the rotor.

Inspired by induction plates

Similar to classical rotary cyclers, the MI-Cycler shines with a very accurate well-to-well temperature uniformity of 0.05°C. However, heating rates of 4°C/s and cooling rates of 3°C/s are merely on average and provide qPCR cycling times of about 25 minutes. The explanation is simple: the rotating aluminium rotor acts similar to a heating block that transfers heat via conduction to the tubes. It is also possible to directly heat metal (iron) dotted PCR tubes through magnetic induction, which would lead to considerably faster ramp rates. According to John Corbett Jr, his company is already working on this technique to cut qPCR cycling times below ten minutes.

Other alternative heating systems already implemented in commercial PCR and qPCR cyclers are based on resistive heating of small disposable plates or silicon wafers. Examples are BJS Biotechnologies Xxpress Cycler and Cepheids Smart Cycler (see *Lab Times* 4/2013). But researchers are continuously tinkering on even smaller and especially more direct heating strategies. One is Laser PCR, developed by the German start-up company GNA Biosolution. The concept of Laser PCR is simple: instead of transferring heat from the reaction tube to the molecules of the PCR reaction mix, heat is directly produced inside the mix by focussing a laser beam on nanoparticles decorated with DNA templates functioning as tiny PCR platforms. After elongation of PCR primers binding to the DNA templates, a laser pulse heats up the nanoparticles to denature the newly synthesised double strands. GNA Biosolutions Laser PCR instrument Pharos is already in the test phase and may be applied for rapid diagnostics of pathogens.

Hot electrons

Photonic PCR, recently proposed by the group of Luke Lee from the University of California in Berkeley is also based on a new heating concept. (Son *et al.*, *Light: Science & Applications* 2015, 4, e280). The photonic PCR cycler utilises LED light to heat a thin gold film in no time at all. As soon as the photons hit the gold surface, a plasmon-assisted light absorption occurs, which in turn excites surface electrons to higher energy states. The excited hot electrons reach temperatures of several thousand Kelvin within 100 femtoseconds – but they also cool down in a few pico seconds, if the light is turned off. Son *et al*. applied this technique to a photonic PCR thermocycler prototype, realising heating rates of approx. 13°C and 7°C, respectively. Needless to say that their ultimate goal is to turn the photonic PCR prototype into a cheap, small, portable thermocycler for molecular diagnostics – that's where the money is.

Harald Zähringer

Thermocyclers

Company/Distributor	Name of Device	Heating Technology	Heating | Cooling Rate ■ Block uniformity	Miscellaneous, Specialities, Generally	Price [EUR]
Agilent Technologies Waldbronn, Germany www.genomics.agilent.com **Contact:** Bernd Martin Phone +49 531 575787 bernd.martin@agilent.com	SureCycler 8800 Thermal Cycler	Peltier	6°C/s for 96 wells | 4°C/s for 384 wells ■ ±0.4°C	Interchangeable 96 and 384 well block | Gradient function: 30–99°C | 10–100 µl working volume | Pre-loaded protocols | High-resolution 7" touchscreen	6.765.– (96 well block)
Analytik Jena Jena, Germany www.analytik-jena.com **Contact:** Melanie Kelm Phone +49 3641 779461 Melanie.kelm@analytik-jena.de	qTower³	Peltier	8°C/s | 6°C/s ■ ±0.15°C (at 55°C after 15 s)	96 well silver sample block in SBS-format with outstanding ramping rates and block uniformity | Open system for plastic ware and chemistry | 10 years long-term warranty on optical system | Flexible and customisable filter module configuration for up to 6-plex measurement | License-free software with all commonly used analysis tools for real-time PCR	19,450.–
	qTower	Peltier	12°C/s | 8°C/s ■ ±0.2°C (at 55°C after 15 s)	Low-profile 96 well silver sample block with highest ramping rates and block uniformity | Minimised reaction volume down to 2 µl with associated plastic ware | 10 years long-term warranty on optical system | Flexible and customisable filter module configuration for up to 4-plex measurement | License-free software with all commonly used analysis tools for real-time PCR	20,450.–
	TOptical	Peltier	6°C/s | 4.5°C/s ■ ±0.15°C (at 55°C after 15 s)	96 well silver sample block in SBS-format with high ramping rates and block uniformity | Open system for plastic ware and chemistry | 10 years long-term warranty on optical system | Flexible and customisable filter module configuration for up to 6-plex measurement | License-free software with all commonly used analysis tools for real-time PCR	19,450.–
	Biometra TOne	Peltier	4°C/s | 3°C/s ■ ±0.20°C (at 55°C after 15 s)	7" colour touchscreen | High Performance Smart Lid (HPSL) for optimal lid pressure and excellent temperature uniformity | Whisper Quiet Technology: Very low noise emission | Open system for plastic ware | Software option: Linear Gradient Tool, Extended Self-Test, remote control via App	On request
	Biometra TAdvanced	Peltier	8°C/s | 5.5°C/s ■ ±0.15°C (at 55°C after 15 s)	7" colour touchscreen | High Performance Smart Lid (HPSL) for optimal lid pressure and excellent temperature uniformity | Fastest ramping – highest average ramping rates due to silver block | Flexible due to Quick-Block-Exchange (60 well/96 silver well / 96 alu well or / 384 well) | Software option: Linear Gradient Tool, Advanced User Management, Protocol Wizard, Extended Self-Test, remote control via App	On request
	Biometra Trio	Peltier	5°C/s | 4.5°C/s ■ ±0.20°C (at 50°C after 15 s)	7" colour touchscreen | High Performance Smart Lid (HPSL) for optimal lid pressure and excellent temperature uniformity | 3 independent blocks | Whisper Quiet Technology: Very low noise emission | Software options: Extended Self-Test, Temperature Optimisation Step, Advanced User Management, Protocol Wizard, User-specific Quick Start, remote control via App	On request
Bibby Scientific Staffordshire, UK www.bibby-scientific.com **Contact:** Phone +44 1785 812 121 info@bibby-scientific.com	Prime Pro 48	Peltier	Average 5.5°C/s ■ ±0.1°C at 95°C	Hollow pure silver block filled with conductive fluid, gold electroplated | 40 cycles in 40 minutes (under 20 minutes optimised) with standard chemistry | Process up to 40% more samples per hour than a traditional 96 well instrument | MIQE guidelines compliant software	On request
	3Prime Thermal Cycler	Peltier	Up to 3.0°C/s ■ ±0.3°C at 55°C	Colour touchscreen user interface for fast program setup | Small space-saving footprint | Block options; 24 x 0.2 ml (compatible with 8 tube strips) and 18 x 0.5 ml | Data transfer via USB | Techne 4 year warranty	On request
	3PrimeX	Peltier	Up to 3.0°C/s ■ ±0.3°C at 55°C	Colour touchscreen interface for fast program setup | Small space-saving footprint | Larger block sizes; 48 x 0.2 ml (half 96 well plate) and 30 x 0.5 ml	On request
	Prime Full Size	Peltier	Up to 3.4°C/s ■ ±0.3°C at 55°C	Colour touchscreen for fast program setup | Upgrade to gradient with a simple unlock code | Versatile block formats	On request
Bio-Budget Technologies Krefeld, Germany www.biobudget-shop.de **Contact:** Karin Widulle Phone +49 2151 6520830 info@bio-budget.de	Labcyler	Peltier	4.2°C/s | 3.6°C/s ■ ±0.25°C at 55°C, ±0.4°C at 95°C	High quality "Made in Germany" | Easily exchangeable, gold-coated silver blocks for different tube formats; 96 well block included | Motor lid with programmable pressure included, ideal for using PCR plates | Space-saving, energy-saving and quiet | Optional gradient update	5,875.–
	Labcycler Gradient	Peltier	4.2°C/s | 3.6°C/s ■ ±0.25°C at 55°C, ±0.4°C at 95°C	Gradient up to 40°C (±20°C) over the entire temperature range | High-quality "Made in Germany" | Easily exchangeable, gold-coated silver blocks for different tube formats; 96 well block included | Motor lid with programmable pressure included, ideal for using PCR plates | Space-saving, energy-saving and quiet	6,575.–
	Labcycler Triple	Peltier	2.5°C/s | 2.2°C/s ■ ±0.25°C at 55°C, ±0.4°C at 95°C	Three independent blocks in one unit; for 3 x 21 tubes at 0.2 ml | Block easily exchangeable | High-quality "Made in Germany" | Motor lid with programmable pressure included, ideal for using PCR plates | Space-saving, energy-saving and quiet	6,920.–

Thermocyclers

Company/Distributor	Name of Device	Heating Technology	Heating | Cooling Rate ■ Block uniformity	Miscellaneous, Specialities, Generally	Price [EUR]
Bio-Budget Technologies (continued) **Contact:** see page 46	Labcycler Compact	Peltier	3.5°C/s | 3.2°C/s ■ ±0.25°C at 55°C, ±0.4°C at 95°C	Block for 48 tubes at 0.2 ml or 48 well PCR plates | Software and operation fully compatible with the 96 well cyclers | High-quality "Made in Germany" | Extremely compact, energy-saving and quiet | Optional gradient update	2,895.–
	Labcycler Compact Gradient	Peltier	3.5°C/s | 3.2°C/s ■ ±0.25°C at 55°C, ±0.4°C at 95°C	Gradient up to 20°C (±10°C) over the entire temperature range | Block for 48 tubes at 0.2 ml or 48 well PCR plates | Software and operation fully compatible with the 96 well cyclers | High-quality "Made in Germany" | Extremely compact, energy-saving and quiet	3,095.–
	Labcycler Compact Silver	Peltier	5°C/s | 5°C/s ■ ±0.25°C at 55°C, ±0.4°C at 95°	Gold-coated 48 well silver block and gradient up to 20°C (±10°C) over the entire temperature range | Software and operation fully compatible with the 96 well cyclers | High-quality "Made in Germany" | Extremely compact, energy-saving and quiet	3,595.–
Bio-Rad Laboratories Muenchen www.bio-rad.com **Contact:** Marcus Neusser Phone + 49 89 31884 177 Marcus_Neusser@ bio-rad.com	T100 Thermal Cycler	Peltier	4°C/s ■ ±0.5°C	96 well: 0.2 ml high-profile | 5.7" graphical touchscreen | Thermal gradient span 1–25°C	5,040.–
	S1000 Thermal Cycler	Peltier	5°C/s ■ ±0.4°C	LCD panel and keypad | Independent blocks: dual 48 well; 0.2 ml | Faster ramping: 96 well; 0.2 ml | Higher volume: 96 deep well; 0.2 and 0.5 ml | High throughput: 384 well	From 7,060.–
	C1000 Touch Thermal Cycler	Peltier	5°C/s ■ ±0.4°C	Upgrades to a CFX96 or CFX384 system | 8.5" graphical touchscreen | Independent blocks: dual 48 well; 0.2 ml | Faster ramping: 96 well; 0.2 ml | Higher volume: 96 deep well; 0.2 and 0.5 ml | High throughput: 384 well	From 8,070.–
	CFX96 Touch Real-Time PCR Detection System	Peltier	5°C/s ■ ±0.4°C	Multiplexing up to 5 targets per well | Detects 1 copy of target sequence in human genomic DNA | Dynamic range: 10 orders of magnitude | Stand-alone system | Automation and LIMS option | Include MIQE annotations and generate RDML files using Biogazelle's qbase+ software	On request
	CFX384 Touch Real-Time PCR Detection System	Peltier	2.5°C/s ■ ±0.4°C	Multiplexing up to 4 targets per well | Detects 1 copy of target sequence in human genomic DNA | Dynamic range: 10 orders of magnitude | Stand-alone system | Automation and LIMS option | Include MIQE annotations and generate RDML files using Biogazelle's qbase+ software	On request
	CFX Connect Real-Time PCR Detection System	Peltier	5°C/s ■ ±0.4°C	Multiplexing up to 2 targets per well | Detects 1 copy of target sequence in human genomic DNA | Dynamic range: 10 orders of magnitude | Stand-alone system | Automation option | Include MIQE annotations and generate RDML files using Biogazelle's qbase+ software	On request
	CFX96 Touch Deep Well Real-Time PCR Detection System	Peltier	2.5°C/s ■ ±0.4°C	10–125 µl reaction volumes | Multiplexing up to 5 targets per well | Detects 1 copy of target sequence in human genomic DNA | Dynamic range: 10 orders of magnitude | Stand-alone system | Automation and LIMS option | Include MIQE annotations and generate RDML files using Biogazelle's qbase+ software	On request
	CFX96 Real-Time PCR Detection System–IVD	Peltier	5°C/s ■ ±0.4°C	10–50 µl reaction volumes | Intuitive CFX Manager IVD software | Multiplexing up to 5 targets per well | Detects 1 copy of target sequence in human genomic DNA | Dynamic range: 10 orders of magnitude	On request
	CFX96 Deep Well Real-Time PCR Detection System–IVD	Peltier	2.5°C/s ■ ±0.4°C	10–125 µl reaction volumes | Intuitive CFX Manager IVD software | Multiplexing up to 5 targets per well | Detects 1 copy of target sequence in human genomic DNA | Dynamic range: 10 orders of magnitude	On request
Biostep Burkhardtsdorf, Germany www.biostep.de **Contact:** Ilona Marzian, Phone +49 3721 3905 0 info@biostep.de	3PrimeBASE Cycler	Peltier	3.0°C/s ■ ± 0.3°C	For 24 x 0.2 ml, 18 x 0.5 ml | 10–100°C (4°C final hold) | 3.5" touchscreen | Can hold 1,000 programmes with unlimited storage available on USB data stick | 4 year warranty	2,495.-
	3PrimeX and 3PrimeG Cycler Gradient	Peltier	3.0°C/s ■ ± 0.3°C	For 48 x 0.2 ml, 30 x 0.5 ml | 10–100°C (4°C final hold) | 3.5" touchscreen | Can hold 1,000 programmes with unlimited storage available on USB data stick | With and without gradient function, gradient range 30–80°C | 4 year warranty	3,645.– / 3,245.– (with/without gradient)
	Prime and PrimeG Cycler Gradient	Peltier	3.4°C/s ■ ± 0.3°C	For 96 x 0.2 ml, 60 x 0.5 ml, 384 well or combi-block (33 x 0.2 ml and 33 x 0.5 ml) | 10–100°C (4°C final hold) | 5.7" touchscreen | Can hold 1,000 programmes with unlimited storage available on USB data stick | With and without gradient function, gradient range 30–80°C | *in situ* hybridisation adapter available | 4 year warranty	4,545.– / 4,045.– (with/without gradient)
	Alpha Cycler Gradient	Peltier	3.4°C/s ■ ± 0.3°C	For 96 x 0.2 ml or 384 well | 10–100°C (4°C final hold | Programme interface: 7" HD Android tablet | Max. gradient 29°C, min. gradient 1°C | 1,000 programmes, data transfer: USB port	4,375.–

Thermocyclers

Company/Distributor	Name of Device	Heating Technology	Heating | Cooling Rate ■ Block uniformity	Miscellaneous, Specialities, Generally	Price [EUR]
Biostep (continued) **Contact:** see page 47	Alpha Cycler Quad block Gradient	Peltier	3.4°C/s ■ ± 0.3°C	For 4 x 96 well or 4 x 384 well blocks | Options: all 96 well or all 384 well, or mixed, separately controllable | 10–100°C (4°C final hold) | Programme interface: 10" HD Android tablet | Clear responsive touchscreen | 1,000 programmes, data transfer: USB port | Input primer sequence	15,000.–
	Real-Time-Cycler Prime Pro 48	Peltier	5.5°C/s ■ ± 0.15°C	For 48 well mini format (1/8 of standard 384 well plates) | hollow silver block, peltier-based-system with conductive fluid | Dramatically reduces the qPCR reagent volumes to a traditional 96 well instrument | Validated for 5–20 µl per well | 30–100°C | With 4 emission filters (505–545 nm, 562–596 nm, 604–644 nm, 665–705 nm) | Open licence software | 400 analyte-specific qPCR reagent kits available: Kits are lyophilised	14,995.–
Biozym Scientific Hess. Oldendorf, Germany www.biozym.com **Contact:** Helmut Prechel Phone +49 5152 9020 support@biozym.com	MIC Magnetic Induction qPCR Cycler	Magnetic induction	6°C/s ■ ± 0.05°C	Outstanding uniformity: ±0.05°C, ideal for HRM and quantifications | 48 samples | 2 or 4 channels | Complete qPCR suite, USB/bluetooth | 15 x 15 cm footprint	Starting from 13,900.–
	LineGene 48+ qPCR System	Ferrotec Peltier	4°C/s ■ ± 0.3°C	48 x 0.2 ml tubes or 8 well strips | Ferroctec high-performance peltiers | 4 channels (LED/PMT) | Motorised lid	On request
	LineGene 96+ qPCR System	Ferrotec Peltier	4°C/s ■ ± 0.3°C	96 x 0.2 ml tubes or plate | Ferroctec high-performance peltiers | 2, 4 or 5 channels (LED/PMT) | Motorised lid | Gradient function	Starting from 16,900.–
	GeneTouch Thermal Cycler	Ferrotec Peltier	4°C/s ■ ± 0.3°C	Interchangeable blocks (single and dual) | 96 well; 2 x 48 well, 384 well, 4 slides | Ferroctec high-performance peltiers | Gradient function (96 well) | Extremely quiet operation	4,990.–
	LifeTouch Thermal Cycler	Ferrotec Peltier	4°C/s ■ ± 0.3°C	96 x 0.2 ml tubes or plate | Ferroctec high-performance peltiers | Gradient function | Extremely quite operation	4,490.–
	LifeECO Thermal Cycler	Ferrotec Peltier	4°C/s ■ ± 0.3°C	96 x 0.2 ml tubes or plate | Ferroctec high-performance peltiers | Gradient function	3,990.–
BJS Biotechnologies Greenford (London), UK www.xxpressPCR.com **Contact:** Ralph Coney Phone +44 203 021 3750 sales@xxpressPCR.com	xxpress	Resistive heating	10°C/s ■ +/-0.3°C	5 colour multiplexing | Only 10% chemistry running costs | Multi-format	19,500.–
Carl Roth Karlsruhe, Germany www.carlroth.de **Contact:** Stefanie Seipp Phone +49 72156061038 s.seipp@carlroth.de	Thermal Cycler 3Prime Personal (Techne)	Peltier	3.0°C/s | 2.1°C/s ■ ±0.3°C at 55°C	Small format for each individual bench-top | 24 x 0.2 ml or 18 x 0.5 ml block | Fast heating and cooling rates plus high temperature homogeneity | Simple, intuitive programming using the 3.5" screen touch interface | All instruments equipped with USB port and oligo-calculator | 4 years warranty on cyclers and blocks	2,687.50 Special prices on request
	Thermal Cycler 3Prime Standard (Techne)	Peltier	3.0°C/s | 2.1°C/s ■ ±0.3°C at 55°C	Standard setting for all general PCR applications, including 48 well plates | 48 x 0.2 ml, 48 well, 30 x 0.5 ml block | Fast heating and cooling rates plus high temperature homogeneity | Simple, intuitive programming using the 3.5" screen touch interface | All instruments equipped with USB port and oligo-calculator | 4 years warranty on cyclers and blocks	3,493.75
	Thermal Cycler 3Prime Gradient (Techne)	Peltier	3.0°C/s | 2.1°C/s ■ ±0.3°C at 55°C	Gradient (30–80°C) with 14°C max. range and gradient calculator | 48 x 0.2 ml tubes or 48 well plates, 30 x 0.5 ml block | Fast heating and cooling rates plus high temperature homogeneity | Simple, intuitive programming using the 3.5" screen touch interface | All instruments equipped with USB port and oligo-calculator | 4 years warranty on cyclers and blocks	3,923.75
	Thermal Cycler Prime Standard (Techne)	Peltier	3.4°C/s | 1.0°C/s ■ ±0.3°C at 55°C	Full-fledged cycler available with mono-blocks for tubes and plates and with combi-block (0.2 ml plus 0.5 ml tubes) | 96 x 0.2 ml tubes or 96 well plates, 60 x 0.5 ml tubes, 384 well plates, combi-block for 33 x 0.2 ml plus 33 x 0.5 ml tubes | Very fast heating and cooling rates plus excellent temperature homogeneity | Simple, intuitive programming using the 5.7" screen touch interface and graphic program surface | All instruments equipped with USB port and oligo-calculator | 4 years warranty on cyclers and blocks	4,353.75
	Thermal Cycler Prime Gradient (Techne)	Peltier	3.4°C/s | 1.0°C/s ■ ±0.3°C at 55°C	Full-fledged gradient-cycler available with mono-blocks for tubes and plates and with combi-block (0.2 ml plus 0.5 ml tubes) | 96 x 0.2 ml tubes or 96 well plates, 60 x 0.5 ml tubes, 384 well plates, combi-block for 33 x 0.2 ml plus 33 x 0.5 ml tubes | Very fast heating and cooling rates plus excellent temperature homogeneity plus 29°C gradient range | Simple, intuitive programming using the 5.7" screen touch interface and graphic program surface | Equipped with USB port and oligo-calculator | 8 elements per block | 4 years warranty on cyclers and blocks	4,891.25

Thermocyclers

Company/Distributor	Name of Device	Heating Technology	Heating | Cooling Rate ■ Block uniformity	Miscellaneous, Specialities, Generally	Price [EUR]
Eppendorf Hamburg, Germany www.eppendorf.de **Contact:** Phone +49 1803 666 789 application-hotline@ eppendorf.de	Mastercyler pro S	Peltier	6°C/s | 4.5°C/s	Vapo-protect technology to reduce evaporation | Network of up to 30 cyclers possible | Gradient span of 24°C | 6 peltier elements	9,106.–
	Mastercyler pro 384	Peltier	4°C/s | 3°C/s	See above	8,327.–
	Mastercyler pro	Peltier	4°C/s | 3°C/s	See above	7,906.–
	Mastercyler nexus GX2	Peltier	3°C/s | 2°C/s	Dual block (64 and 32 wells) | 64 well block with gradient | Up to 3 units can be combined | 6 peltier elements	9,100.–
	Mastercyler nexus X2e	Peltier	3°C/s | 2°C/s	Dual block (64 and 32 wells) | Controlled through any another Mastercyler nexus | Up to 3 units can be combined | 6 peltier elements	5,800.–
	Mastercyler nexus GSX1	Peltier	5°C/s | 3.5°C/s	Flexlid for flexible use of consumables | Gradient span of 20°C | Up to 3 units can be combined | 6 peltier elements	8,500.–
	Mastercyler nexus SX1e	Peltier	5°C/s | 3.5°C/s	Flexlid for flexible use of consumables | Controlled through any another Mastercyler nexus | Up to 3 units can be combined | 6 peltier elements	5,700.–
	Mastercyler nexus flat	Peltier	3°C/s | 2°C/s	Flat block for *in-situ* PCR and other formats | 6 peltier elements	7,200.–
	Mastercyler nexus gradient	Peltier	3°C/s | 2°C/s	Flexlid for flexible use of consumables | Gradient span of 20°C | Up to 3 units can be combined | 6 peltier elements	7,900.–
	Mastercyler nexus eco	Peltier	3°C/s | 2°C/s	Flexlid for flexible use of consumables | Controlled through any another Mastercyler nexus | Up to 3 units can be combined | 6 peltier elements	4,995.–
Inheco Martinsried, Germany www.inheco.com **Contact:** Andreas Scholle Phone +49 89 899593 101 AScholle@inheco.com	On Deck Thermal Cycler 96 – ODTC 96	Peltier, Vapor Chamber-VCM	Average 4.4°C/s | 2.2°C/s ■ ±0.2°C	Only for integration on liquid handling workstations	On request
	On Deck Thermal Cycler 384 – ODTC 384	Peltier, Vapor Chamber-VCM	Average 4.4°C/s | 2.2°C/s ■ ±0.2°C	Only for integration on liquid handling workstations	On request
LTF Labortechnik Wasserburg, Germany www.labortechnik.com **Contact:** Noel Kändler Phone +49 8382 9852 0 info@labortechnik.com	MyGo Pro	Peltier	5°C/s | 4°C/s ■ 0.05°C (SD)	HRM | Full spectrum optics (FSO) | 12 channels | 0.1 ml tubes/8-tube strips | 32 well, expandable to 96 and more	From 14,500.–
	MyGo Mini	Peltier	3°C/s | 1.5°C/s ■ 0.05°C (SD)	HRM | Portable (weight under 2 kg) | Noiseless | 16 x 0.1 ml | 3 channels	From 8,500.–
	Q-Cycler 24	Peltier	Up to 2.5°C/s | Up to 2°C/s ■ < 1°C	24 x 0.2 ml | Colour touchscreen	From 2,790.–
	Q-Cycler 48	Peltier	See above	48 x 0.2 ml | Colour touchscreen	From 3,390.–
	Q-Cycler 96+	Peltier	Up to 6°C/s | Up to 3°C/s ■ < 1°C	96 x 0.2 ml | Colour touchscreen | TAS system included | Different blocks available	From 6,990.–
miniPCR Cambridge, MA, USA www.minipcr.com **Contact:** Sebastian Kraves Phone +1 781 990 8727 sales@minipcr.com	MiniPCR Mini8 Thermal Cycler	Resistive heating, forced air cooling	3°C/s | 2°C/s (max) ■ 0.1°C	Portable: 2 x 5 x 4" | Weighs less than 1 lb (0,373 kg) | Compatible with Li-Ion batteries | Smartphone and computer operable | Designed and made in USA	585.–
Nippon Genetics Düren, Germany www.nippongenetics.eu **Contact:** Marcelo Lanz Phone +49 2421 554960 info@nippongenetics.eu	Kyratec Super-Cycler Trinity for Endpoint PCR	Peltier	7°C/s ■ ±0.3°C	Three temperature zones | Temperature range of 4–99°C | 7" touchscreen | Touch down/up function | Wizard mode for quick set up | Block with 3 fully independent sets of peltier elements |	On request
	Kyratec Super-Cycler Unity for Endpoint PCR	Peltier	7°C/s ■ ±0.3°C	Affordable | Temperature range of 4–99°C | 7" touchscreen | Touch down/up function | Wizard mode for quick set up | Block with peltier elements	On request
Qiagen Hilden, Germany www.qiagen.com **Contact:** Phone +31 0800 0229592 orders-bnl@qiagen.com	Rotor-Gene Q	Air heated rotary cycler	-- ■ ±0.02°C	Uniform detection eliminating the need for ROX reference dye | Fast ramping and negligible equilibration times for short run-times | Complete confidence in your results	On request
Roche Diagnostics Roche Diagnostics www.lightcycler.com **Contact:** lifescience.rms@roche.com	LightCycler 480 II System	Peltier with Therma-Base	4.4°C/s | 2.2°C/s (96 well) 4.8°C/s | 2.5°C/s (384 well)	Therma-Base technology | 20 channels/ 6-plex multiplexing | 96/384 well exchangeable block | Instant detection (Scan free) | Proven robustness	On request
	LightCycler 96 System	Peltier	4.4°C/s | 2.2°C/s	Fast electro-formed block technology | 4 channels / 4-plex multiplexing | 96 well block | Instant detection (Scan free) | Intuitive touchscreen	On request

Thermocyclers

Company/Distributor	Name of Device	Heating Technology	Heating | Cooling Rate ■ Block uniformity	Miscellaneous, Specialities, Generally	Price [EUR]
Roche (continued) **Contact:** see page 49	LightCycler 1536 System	Peltier with Therma-Base	4.8°C/s | 2.5°C/s	Therma-Base technology | 2 channels/ 2-plex multiplexing | 1,536 well block | Instant detection (Scan free) | Automation compatible	On request
SensoQuest Goettingen, Germany www.sensoquest.de **Contact:** Kay Terpe Phone +49 17666646603 k.terpe@sensoquest.de	Labcycler Basic & Labcycler Gradient	Peltier	4.2°C/s | 3.6°C/s (50–99°C) ■ ±0.25°C at 55°C ±0.40°C at 90°C	Self-calibrating system | Touchscreen: TFT illuminated colour display | Low energy consumption | Fully automatic lid: Temperature and pressure programmable, electric moving | Electroformed gold coated silver blocks (48, 96, 384) | Thermal conductivity: 429 W/mK	Please contact your local distributor
	Labcycler Triple	Peltier	2.5°C/s | 2.2°C/s (50–99°C) ■ ±0.25°C at 55°C ±0.40°C at 90°C	3 total independent PCR runs in one system | Condensing protection with passive lids | Three small aluminium blocks (3 x 21, 0.2 ml) | Thermal conductivity: 237 W/mK	Please contact your local distributor
	Labcycler 48 & Labcycler 48 Gradient	Peltier	3.5°C/s | 3.2°C/s (50–99°C) ■ ±0.25°C at 55°C ±0.40°C at 90°C	Small version of the Labcycler | TFT touchscreen | Gradient capable | Program transfer between Labcycler Basic and Labcycler 48 possible | Small aluminium blocks (0.2 ml tubes) | Thermal conductivity: 237 W/mK	Please contact your local distributor
	Labcycler 48s & Labcycler 48s Gradient	Peltier	5.0°C/s | 5.0°C/s (50–99°C) ■ ±0.25°C at 55°C ±0.40°C at 90°C	Highest speed of all Labcyclers | Gradient capable | TFT touchscreen | Administrator protection | Small silver blocks (0.2 ml tubes) | Thermal conductivity: 429 W/mK	Please contact your local distributor
Thermo Fisher Scientific www.thermofisher.com/contactus	Applied Biosystems SimpliAmp Thermal cycler	VeriFlex technology	4°C/s ■ <0.5°C (30 s after reaching 95°C)	Large, responsive touchscreen | Thermal simulation modes – simulate older cyclers and avoid re-optimising your programs | Precise temperature control with Veriflex technology | Cloud connectivity	4,990.–
	Applied Biosystems ProFlex PCR system	VeriFlex technology	6°C/s ■ <0.5°C (30 s after reaching 95°C)	Multi-user: run three separate PCRs at the same time | Thermal simulation modes – simulate older cyclers and avoid re-optimising your programs | Flexible configuration – 5 interchangeable block formats to choose from | Cloud connectivity	8,770.– to 11,350.– (depending on block format)
VWR International Erlangen, Germany https://de.vwr.com **Contact:** Christof Larisch Phone +49 9131 6107020 info.peqlab@de.vwr.com	UNO96 Thermocycler with 96-well universal block	Peltier	5°C/s ■ ±0.2°C (at 72°C)	16 Peltier elements | 96 well thermocycler suitable for medium and high sample throughput | Innovative emulation mode | Reliable, reproducible results | Highest quality | Free PCR Cycler Master PC software	4,980.– (please ask for country specific prices)
	UNO96 Gradient Thermocycler with 96-well universal gradient block	Peltier	5°C/s ■ ±0.2°C (at 72°C)	16 Peltier elements | 96 well thermocycler suitable for medium and high sample throughput | Innovative emulation mode | Reliable, reproducible results | FlexGradient Technology for a perfect linear gradient | Free PCR Cycler Master PC software	5,340.– (please ask for country specific prices)
	UNO384 HPL Thermocycler with 384-well block and High Pressure Lid	Peltier	5°C/s ■ ±0.2°C (at 72°C)	16 Peltier elements | 384 well thermocycler for highest sample throughput equipped with high pressure lid technology to avoid evaporation | Reliable, reproducible results | Simple to use | Highest quality | Free PCR Cycler Master PC software	6,000.– (please ask for country specific prices)
	Doppio Thermocycler with 2x 48-well universal blocks	Peltier	5°C/s ■ ±0.2°C (at 72°C)	2 x 8 Peltier elements | Two independent 48 well block thermocyclers in one system, for a maximum of flexibility | Innovative emulation mode | Reliable, reproducible results | Highest quality | Free PCR Cycler Master PC software	5,490.– (please ask for country specific prices)
	Doppio Gradient Thermocycler with 2x 48-well universal gradient blocks	Peltier	5°C/s ■ ±0.2°C (at 72°C)	2 x 8 Peltier elements | Two independent 48 well block thermocyclers in one system, for a maximum of flexibility | Innovative emulation mode | Reliable, reproducible results | FlexGradient Technology for a perfect linear gradient | Free PCR Cycler Master PC software	5,590.– (please ask for country specific prices)
	Ristretto Thermocycler with 32-well universal block	Peltier	3°C/s ■ ±0.2°C (at 72°C)	8 Peltier elements | Compact personal cycler with highest flexibility | Reliable, reproducible results | Simple to use | Highest quality | Free PCR Cycler Master PC software	2,440.– (please ask for country specific prices)
WaferGen Biosystems Val Fleuri, Luxembourg www.wafergen.com **Contact:** Marizela Kulisic Phone +352 26 970 970 info.europe@wafergen.com	SmartChip MyDesign Real-Time PCR System	--	--	Includes SmartChip MultiSample NanoDispenser and SmartChip Real-Time PCR Cycler | Also includes analysis software with 5 user licenses	154,140.–

Tips and tricks of the trade

DNA Origami Comes of Age

It's been just ten years since Paul Rothemund found a simple way to generate nanoscale DNA structures similar to paper origamis. Today, DNA origamis may help to construct nanomachines or deliver DNA into cells.

Lab Hint

Someone I know uses a laptop as a book end. "That's not what it is for," I explained. "It is meant to be a sophisticated information storage and processing device, and you are using it as a piece of construction." That is what has been happening lately with DNA. As far as most of us are concerned, DNA uses base-pairing for the purpose of replication and the fact that it also holds the two strands together is almost a side-effect.

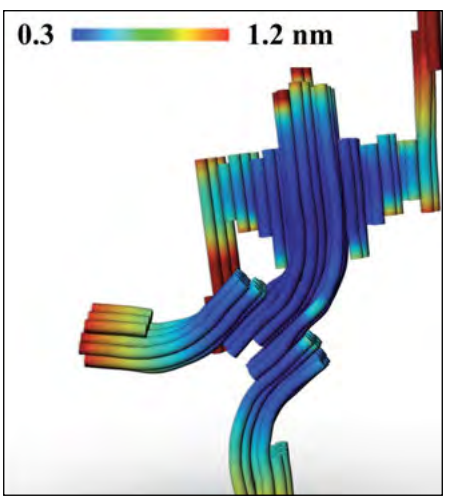
Rightly so, perhaps. But some labs have taken their lead from my friend and have decided to misuse this elegant information storage device and hijack it for quite different purposes. Instead of using base-pairing to copy information, they have used it to tangle DNA up into sheets and tiles, to make tiny structures and even machines. It is called DNA origami. And it all began with the Holliday knot. Back in the deep mists of time (the 1960s) a British geneticist called Robin Holliday worked out a crazy variation of homologous recombination. We are all familiar with the way two strands can recombine using AT/CG pairing but Holliday suggested that something odd might happen, when you force not two but four strands to pair up. He proposed that if you do that, you could end up with a four-arm structure.

Jumping game

How can that happen? Well, first imagine you are zipping up strands – sticking strand A, say, onto strand B. Now imagine we add a third strand, C, complementary to A. As A gets stuck onto B, it "jumps" onto strand C at some point. At that very point, the jilted strand B pairs up with a fourth strand D, complementary to B. Result: a cross-shaped, four-armed structure – the Holliday junction.

Our genetically-minded, cell-cycle aware readers will point out that this is nothing special and, indeed, the Holliday junction is a precursor to many types of genetic recombination. But where DNA origami comes in, is where you realise that you can play the jumping game in different ways. Jump once and you get a Holliday junction. Keep on jumping in a regular pattern and you get a woven sheet, or tile, of DNA. It is a bit like weaving with DNA, using base-pair complementarity to stick the fibres together.

DNA origami is usually done by taking a single-stranded genome of the M13 bacteriophage. This is a big loop that you are going to fold up into the shape you want. You



DNA robot designed with the DNA origami programme CanDo.

then add strands of DNA called "staples" because they staple the loop into shape. Thanks to complementarity, the DNA staples automatically tangle up in a predictable way, to give you the shape you want.

And there is no limit to that shape – you can have tubes, poles, plates, whatever. Most often, you will build up your final structure by making standard shapes (such as tiles) and stacking them together into something more complicated (like cubes). Paul Rothemund, the inventor of DNA origami, made circles, triangles and even smiley faces at the 100 nm scale (*Nature* 440: 297–302), while Castro *et al.* (*Nat Methods* 8: 221–9) even made model robots, just 75 nm tall.

But how is it done in practice? It is surprisingly easy, once you know how. Or rather, once you have loaded the software. Programmes like CanDo (*http://cando-dna-origami.org*) allow you to specify the shape you want and it will tell you the sequences of DNA staples you need to mix up with scaffold, to get your nanostructure. You then synthesise your staples, add them to your M13 genome and start the annealing reaction. Simple! At least, with a bit of tweaking the conditions. Okay, a lot of tweaking.

What is it good for?

But other than making tiny models of R2D2, what is the point? Actually, DNA origami could turn out to be useful in a whole host of ways. For one, they are a good way of getting DNA into cells without electroporation, because, when the assemblies stick to the surface of cells, they provide a really high local concentration of DNA. Even better, you can make nanomachines that will physically deliver the DNA into the cell, like a tiny drill.

And once you realise you can make just about any shape you want, the imagination starts to run wild. This March, Yonggang Ke *et al.* (*Nature Comms*. 7: 10935) described how they had made a DNA actuator out of four rigid arms of DNA joined into a rhombus. Two arms are joined by an adjustable strut made of a mixture of scaffold and staples, whereas the other two are loaded with the cargo molecule, in this case two fragments of green fluorescent protein. By adding just the right staples, they could open or close the rhombus and control the distance between the two cargo molecules (as measured by fluorescence intensity).

DNA origami, once the fine-tuning has been done, offers a phenomenally quick way of making nanoscale machines. If anyone finds a way of adding controlled movement, the future of this technique will look very exciting indeed.

Steven D. Buckingham

Do you have any useful tips?
Contact us at:
editors@labtimes.org

Bench philosophy (62): Carbon dots from beer

Cheers to New Imaging Tools

Carbon quantum dots offer a new way to fluorescently label living tissues and deliver drugs or genetic material. They are photo-stable, have low toxicity and are highly versatile – and you can even find them in your beer.

Recent years have seen a growth in the deployment of carbon nanomaterials such as fullerenes, nanotubes and graphite carbon dots. Carbon quantum dots (CQDs) are among the simplest of these nanomaterials. CQDs are quasi-spherical nanoparticles of carbon, some 10 nm or less in diameter. When looking a little closer, you will notice they can take on all sorts of appearances, ranging from nearly amorphous lumps to nanocrystalline structures. These odd materials bring some interesting chemical and physical properties, and life scientists are catching on to a range of applications, for which they may be used. With a little bit of modification, you can use carbon quantum dots as biosensors, contrast agents and drug delivery vehicles. You can apply them to monitor how a drug is making its way through a patient's body and you can exploit them to monitor the incorporation of individual nucleotides into an extending DNA strand. Or you can simply use them as a biologically inert, non-toxic, water-soluble contrast agent that doesn't fade during illumination.

Making them can be complex, involving some arcane chemistry and clever tricks with lasers. Or you can just extract them from cornflakes, biscuits and even beer. Yes, I am being serious.

Tiny lumps of charcoal

CQDs are a member of a class of carbon-based nanomaterials. Other examples are graphene – huge hollow sphere, tube or ellipsoid of carbon atoms – and the better-known carbon nanotubes. CQDs can take on several different structures, depending on how you make them but usually they are amorphous – just tiny lumps of charcoal. What makes them interesting is their unusual properties. One such odd property is their fluorescence, even though it is not entirely clear what triggers it. And what is more, the wavelength of the signal they emit is tuneable, as it changes with the wavelength of excitation. This is a major advantage, especially when you are trying to work with more than one fluorescent probe at the same time.

This plastic bag filled with Tsingtao beer should yield enough carbon dots for your fluorescent labelling experiments.

CQDs also have some very useful absorbance properties. This is because they have many C=C bonds, whose π-π^* transitions absorb short-wavelength light. As I explain below, this means that you can attach a light-emitting probe to the CQD and the dot will absorb the signal until the probe becomes detached. Given you some ideas already? I'll let you read about these in a moment but to give you something to get going on, CQDs have some weird properties that will get our more imaginative readers very excited. For example, CQDs are electrochemiluminescent – pass a current through them and they glow. Possible new voltage probes, perhaps?

Bottom-up or top-down?

How do you make CQDs? There are two basic methods – "bottom-up" or "top-down". Making them from the bottom up means finding a chemical synthetic pathway that puts the carbons together in the right pattern. Making them top-down means taking some larger-scale molecular structure and breaking it up, so that you end up with CQDs. Let's look at that a bit more closely. One bottom-up way of making CQDs is to use pyrolysis or carbonisation of organic precursor molecules. Pyro what?, you may ask. Okay, let me start that again. One way of making CQDs is to melt certain compounds (such as amino acids or sugars) until they go brown. Or you can just put sugar in the microwave: several groups have reported good yield of green fluorescent CQDs after just a few minutes' microwave irradiation of a mixture of sucrose and diethylene glycol. Other workers have used electrochemical ablation, where a current is passed through a mixture of precursor compounds.

Crude methods

So what kinds of compounds can you start with? The list includes organic salts, such as ascorbic acid, glutamic acid, glycerol and coffee. Yes, coffee. It gets weirder...

So much for bottom-up approaches, what about the top-down ones? The basic idea is to take some carbon and break it up. Many top-down approaches to making CQDs start off with carbon nanotubes, graphite rods, carbon fibre and even soot. One method is to oxidise the starting material at low pH, although this nearly always introduces a negative charge onto the CQDs, which makes them insoluble, plus it can be difficult to completely remove the oxidising agent. Alternatively, you can cleave the carbon precursors electrochemically, in which case it is thought that the exfoliation into dots is caused by electrochemical stress. This method is simple and has a high yield.

If these methods often seem a bit crude, you are right. The devil is in the detail and the trickiest part is making sure you end up with a homogenous product. Many workers use standard purification techniques, such as ultrafiltration, centrifugation and chromatography. But there are some exciting high-tech ways, too. For instance, one group (Shen *et al*, 2012, *Chem. Commun.* 48: 3686) used a three-step process involving nanoreactors (spherical structures that grab hold of target molecules on their surface). Sticking the organic precursor onto

the nanoreactor is the first step, followed by a pyrolysis step (baking, in other words). Finally, the nanoreactor is dissolved away to release the CQDs.

Let's get Nature to do the work

Does all this sound a bit tricky and technical to you? Okay, then let's get Nature to do the work for us. And this is where things get even odder. It turns out that there are ready-made CQDs in coffee, fizzy drinks and even beer. Mingquian Tan's group at the Dalian Institute of Chemical Physics, China, has discovered a niche role in finding ready-made CQDs in all sorts of strange, yet familiar, places. First of all, they found them in Nescafé, a brand of instant coffee. Then they decided to look for them in Tsingtao beer, a popular brand in China, and went on to show that having extracted them, by first stirring to remove gas followed by evaporative condensation and purification on Sephadex gel, they could use them to show up cancer cells and even deliver anti-cancer drugs. The original paper has an interesting account of the history of Tsingtao and a methods section reminiscent of a cocktail recipe (Wang *et al.* 2015, *Analytical Methods* 7:8911).

Tan went on to show there are CQDs in several soft drinks and it is emerging that they are lurking in just about anything that involves carbonisation, such as bread, caramel and corn flakes.

Once you have got your dots, then the real fun begins. In this case, the fun consists of thinking up things to do with the surface of the dots, to make them do interesting or clever things. This is made easy with CQDs because they have oxygenated functional groups on their surface, which makes them convenient hydrophilic handles, onto which you may stick different chemicals. These materials can be used to alter the native fluorescence of the dots or they can be used in more sophisticated ways, such as delivering drugs or turning the dots into biosensors. And don't forget the dots are non-toxic, so this works for *in vivo* imaging, too.

Surface modifications

Some modifications of the surface are designed just to increase the quantum yield of the dots. The most common is passivation using polyethylene glycol, although this has some negative side effects in that it can make it more difficult to add other chemical moieties and can also increase the size of the dots. Other modifications are just designed to change the colour of the emission, and a body of folklore gradually accumulates to act as a guide as to what to add to get the colour and shade of your choice. For instance, adding zinc sulphide or PEG can alter the emission peak from 510 to 650 nm, meaning you can use simultaneous multiple probes. Remember also that the emission of CQDs changes with the wavelength of the excitation, so you have a lot of room to manoeuvre to get the right contrast conditions.

CQDs can be used to make very sensitive probes for chemical species. You can passivate dots using branched polyethyleneimine (BPEI), instead of polyethylene glycol, and make yourself a probe for Cu^{2+} that can detect down to 6 nM. It is thought that the copper's interaction with the amino groups of the BPEI quenches the fluorescence of the dots. But this makes for big dots that won't get into cells, although you can get round this by using alternative routes of synthesis.

CQDs have been used to make a pH sensor. One group accomplished this by attaching an established proton receptor (its name is very long, so we won't waste paper with it here) to dots and found the intensity was linear with pH over a wide range, enabling them to read pH in the cytosol in real time (Kong *et al.* 2012, *Adv. Mater.* 24: 5844).

Different applications

CQDs can be used as drug delivery vehicles. This illustrates interplay between the "native" advantages of dots (their biological inertness, high fluorescent yield and photostability) and the "add on" features supplied by the surface modifications. Several labs have confirmed that loading the anticancer drug, doxorubicin (DOX) onto CQDs is effective at selectively killing cancer cells.

Minqian Tan (of Tsingtao beer fame) made CQDs using a simple hydrothermal approach – they baked citric acid and o-phenylenediamine. These dots had (as is common) a negative surface charge, which meant it was trivial for them to adsorb the DOX molecules. But there is an extra feature that comes with this arrangement: as well as delivering the drug to the target cells, the presence of the DOX quenched the dots' fluorescence. This meant that release of the drug could be monitored as the recovery of fluorescence (*Biotechnol Lett* DOI 10.1007/s10529-015-1965-3).

In a similar approach, Zheng *et al.* used another anticancer agent (oxidised oxaliplatin) and were able to track the cellular trafficking of the dots to their target (2014, *Adv Mater* 26: 3554).

But CQDs are not only good for delivering drugs, they can deliver genes too. CQDs modified with polyethylenimine are negatively charged and this is enough to hold DNA molecules for delivery into cells. Qing Wang and colleagues successfully delivered short-interfering RNA against survivin into the human gastric cancer cell line MGC-803, knocking gene expression down by nearly 94% (*J Nanobiotechnol*, 2014 12:58). And remember, you can modify the dots, enabling you to monitor the delivery in the same way as monitoring drug delivery – all in real time, of course.

Many advantages

Carbon dots offer quite a few advantages over "traditional" fluorescent probes. For one, they are astonishingly photo-stable, in contrast to the notorious bleaching of other types of fluorescent probe, which bedevils high-resolution *in vivo* imaging. Whereas traditional organic fluorophores bleach within minutes, CQDs typically lose only about five percent of their fluorescence, even after four hours of irradiation.

They are also very non-toxic. After all, they are just lumps of carbon, the stuff from which we are made. Survival rates in the 90% region have been reported for many cells and tissues, including human hepatocellular carcinoma cells, human breast cancer cell lines and even whole animals. But of course, if they are really going to be useful as drug delivery agents, we have to be sure they will clear from the human body quickly; animal studies have indeed shown this to be the case. Of course, safety concerns go beyond mere toxicity but it is encouraging to note that CQDs have been fed in large quantities to mice and rats without any perceptible change in food intake, body weight, behaviour, kidney or liver function.

With this combination of low toxicity and high photo-stability, coupled with their adaptability to a range of applications, CQDs have the potential to become an important technique in the coming decade. You may have some difficulty, however, explaining why you have ordered Tsingtao beer on the lab budget...

Steven D. Buckingham

Fancy composing an installment of "Bench Philosophy"?

Contact Lab Times
E-mail: *editors@labtimes.org*

New Products

Mixing

Product: Vortex mixer
Name & Manufacturer:
SA6 from Bibby Scientific
Technology: The robust, die-cast-constructed vortex mixer features a sophisticated inbuilt counterbalance system as well as a suction cup feet to prevent any walking issues, which are often associated with budget vortex mixers.
Advantages: The mixing speed ranges from 2500 to 4500 rpm, making it suitable for a range of applications.
More Information: www.bibby-scientific.com

Liquid Handling

Product: Micropipette
Name & Manufacturer: Evolve from Integra Biosciences
Technology: Unlike traditional pipettes, which utilise a single rotating plunger to set volumes, the new model features three adjustable dials for setting each individual volume digit. Simply depressing and twisting the plunger unlocks the volume dials. Once unlocked, the three dials can be freely adjusted to rapidly set the desired volume. This approach allows users to set volumes more than ten times faster.
Advantages: Available in single, eight and twelve channel formats, covering a volume range of 0.2 – 5,000 µl. The ultra-lightweight, well-balanced design of the pipette enhances productivity and comfort even during prolonged pipetting sessions.
More Information:
www.integra-biosciences.com

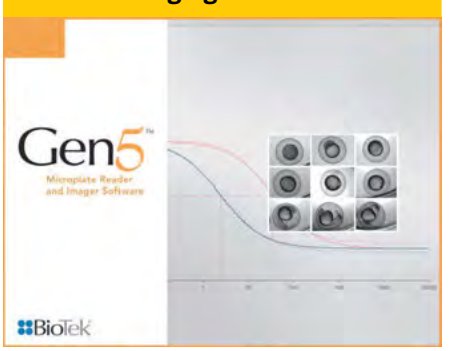
Image Analysis

Product: High-Content Imaging System
Name & Manufacturer: ImageXpress Micro 4 from Molecular Devices
Technology: The modular configuration allows researchers to customise the system to fit individual needs. It now includes the ability to add confocal capability with the AgileOptix spinning disc confocal module without disruption to existing workflows.
Advantages: The system is a premiere, integrated and scalable toolset for optimal 3D image acquisition and analysis.
More Information:
go.moleculardevices.com/ixm-4

Particle Characterisation

Product: Analytical Ultracentrifuge (AUC)
Name & Manufacturer: Optima from Beckman Coulter Life Sciences
Technology: The AUC allows molecules to float free and unbound so that researchers are able to characterise them in their native state. This technology determines molecular weight, size, shape and polydispersity, and is capable of identifying interactions between particles in a native, matrix-free environment. Wavelength-specific monitoring allows researchers to study complex systems in a single experiment.
Advantages: A 38.1 cm (15 inch diagonal) touch-screen display eases operation and boosts efficiency. In addition, modern, intuitive software enables simple run-monitoring and data exports performed either locally or remotely. The optics are contained outside the rotor chamber, making it easier to clean, and reducing the impact of the g-force on the optics.
More Information: info.beckmancoulter.com/OptimaAUC

Live Cell Imaging



Product: Data analysis software
Name & Manufacturer: Gen5 3.0 from BioTek
Technology: Image analysis capabilities are available in two editions, with the new Gen5 Image Prime offering advanced image analysis tools, including cell segmentation and object-level measurements that are critical to many cell-based imaging applications. 21 CFR Part 11 compliant features are also available to meet GxP requirements.
Advantages: The programme offers automated image capture and analysis, real-time annotation tools and built-in time-lapse video production.
More Information: www.biotek.com

Super-Resolution Microscopy

Product: Desktop microscope
Name & Manufacturer: Nanoimager from Oxford Nanoimaging
Technology: The microscope unit is just 21 cm x 21 cm x 15 cm. It has been engineered from the bottom up for optimum single-molecule imaging functionality. It may be operated on a regular desk or bench without the need for extra anti-vibration or environmental isolation. The inherently robust design works together with passive dampening elements to reduce vibrations and drift, and real-time focus and sample positioning provides the stability for data collection over many hours.
Advantages: The clear and intuitive user interface helps researchers new to single-molecule localisation work toward rapid productivity. The large field of view, real-time data analysis features, and high degree of instrument automation enable high-throughput workflows, directly applicable to the wide range of emergent single-molecule screening applications.
More Information: www.oxfordni.com

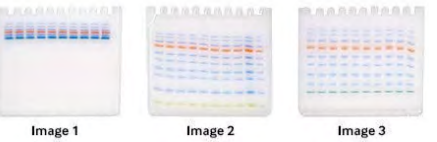
Microbial Fermentation

Product: Rigid-wall single-use vessel
Name & Manufacturer:
BioBLU 3f from Eppendorf
Technology: Robust magnetic overhead drives featuring Rushton-type impellers support agitation rates up to 1200 rpm and provide high-performance mass transfer. Integrated cooling baffles enable efficient heat removal for exothermic processes. The rigid-wall industrial stirred-tank design ensures scalability and simplifies technology transfer. The monolayer polymer material mitigates risk and uncertainty with regard to leachables and extractables.
Advantages: The single-use bioreactors are designed as drop-in replacements for existing autoclavable fermentation vessels.
More Information:
www.eppendorf.com /BioBLUf

Stem Cell Culture

Product: Cryopreservation medium
Name & Manufacturer:
CryoStem from Biological Industries
Technology: The defined, protein-free formulation contains no serum, but rather methylcellulose and DMSO. It has been extensively validated for cryopreservation with human embryonic cells (H1, H9 and HuES9) and iPSC. Suitable for feeder and feeder-free conditions and developed for cell passaging as aggregates.
Advantages: Upon thawing and plating, a very high percentage of viable hESC are obtained with high recovery efficiency. Human embryonic cells show excellent attachment, growth performance, and maintenance of pluripotency markers after thawing with superior results compared to both serum-containing freezing media and other serum-free solutions. Combined with serum-free, xeno-free media, it allows cells to be kept in defined, reproducible and set conditions throughout culture.
More Information:
www.bioind.com/cryostem-freezing-medium

Electrophoresis



Product: Tris SDS PAGE Running Buffer
Name & Manufacturer: FastRun from Fisher Scientific
Technology: Tris-Glycine mini-gels (precast or home-made) prepared with the conventional buffer system are typically run at 125 V. The buffer system heats up at high voltages, which in turn heats the gel, resulting in a breakdown of the protein bands and loss of resolution. The FastRun buffer can be run at a higher voltage (200 V recommended) because it does not generate excessive heat. The result is a significant improvement in run times. In an example, gels run with the new buffer system took only 25 minutes, compared to 90-minute run times with the traditional reagents. The buffer is compatible with standard Tris-Glycine polyacrylamide gels (precast or home-made) and with all commercially available protein electrophoresis tanks.
Advantages: The buffer system provides comparable or better resolution and an increased molecular weight separation range of proteins when compared to traditional Tris-Glycine-SDS buffer. It also reduces the number of Tris-Glycine polyacrylamide gel compositions required to resolve a protein.
More Information:
www.fishersci.com/fastrunsdsbuffer

Cell Culture

Product: Confluency checker software
Name & Manufacturer:
CKX-CCSW from Olympus
Technology: The software utilises an exclusive cell counting algorithm, quickly creating quantifiable cell growth data to determine when cells require passaging, experimentation or storage. Measurement records can be saved and exported as a CSV file for further analysis or archiving.
Advantages: Without having to remove cells from the vessel, the software quantifies the exact growth density.
More Information:

The harlequin ladybird beetle (*Harmonia axyridis*) occurs in numerous colours and with between none and 22 spots.

Photo: Entomart

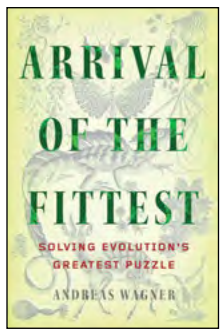
Book Review: Solving evolution's greatest puzzle

Following Prepared Trails

Andreas Wagner shows how evolution depends on the sheer number of possible genotypes. Can this bioinformatician live up to Richard Dawkins as a writer?

How did the bewildering diversity of life on earth arise? "Random mutation and natural selection" is the standard answer that every college graduate would give. Apart from creationists, few would contradict this simple formula of evolutionary biology.

Andreas Wagner from the University of Zurich, however, raises serious objections. The evolutionary biologist shares at least one concern with creationists: How could there have been enough time for organisms to try out all the possibilities? In the prologue of his book, *Arrival of the Fittest*, he discusses the number of possible protein sequences: There are 10^{130} possible proteins made of just 100 amino acids, that is 10^{40} as many arrangements as there are hydrogen atoms in the whole universe. That's in fact a "hyperastronomical" number!



Wagner – far from being a creationist – does not deny the amazing force of natural selection, "But given the staggering odds, selection is not enough. We need a principle that accelerates innovation." Random mutation, for him, needs explaining. And that is, at least partially, what he does in his book.

Random mutation needs explaining

To illustrate his case, he uses the analogy of the universal library that contains all the books that could possibly be written. In it, of course, almost everything is complete gibberish but, "if you wandered through the universal library of books long enough, you would find books that surprise you. They contain novel thoughts, ideas and inventions." Interested readers should start to spot the parallels with evolution about now.

In this boring universal library, each book sits between two neighbours. In the strange library of all possible proteins composed of 100 amino acids, each protein has 1,900 neighbours. So, one neighbour stands for one possible substitution of one amino acid, 19 possibilities at each of the 100 positions. In his bioinformatics lab, Wagner's collaborators make algorithms to walk through such strange hyperastronomical libraries – substitution by substitution from neighbour to neighbour, as evolution does.

Small steps for evolution

Even if only one protein in 10,000 is able to fold, there are still 10^{126} left to perform the magic of life. Four in six billion, or as many as 10^{93} bind ATP. There really is no shortage of possible ATP binders. Different functions yield similar numbers. And the sequences for the same function vary enormously. Take globin-folds that, in extreme cases, share only 20 percent of their sequence. So, there are unimaginably numerous ways to arrive at the same function or fold in the library. And the overwhelming majority of proteins have never been tried by an organism, many became extinct and only very few exist today.

Wagner's libraries are not only hyperastronomical in size but hyperdimensional in structure (1,900 dimensions for 100 amino acid proteins instead of one dimension for books). In this weird space, Wagner's collaborators showed that there are paths – substitution by substitution or neighbour by neighbour – from one end of the library to the other without a change in function. In contrast, it is possible to reach quasi full diversity – most possible functions – in a few steps from each position.

Wagner concludes, "The astonishing fact that evolution needs to explore only 10^{100}th of a library to secure the arrival of the fittest goes a long way to explain how blind search produces life's immense diversity." And proteins are only a subset: Wagner begins with metabolism, where each genotype is characterised as a set of possible biochemical conversions (fructose to glucose, for example). But there are also regulation networks (like hox genes) and RNA.

Reuniting neutralists and selectionists

Complexity allows for flexibility. For Wagner, the characteristics of these libraries resolves the debate between neutralists, who think that most genetic variants are irrelevant, to fitness and selectionists, for whom this would stop the evolutionary process. The paths with the same function within "genotype networks" (as Wagner calls them), "are crucial to find each change, and natural selection is crucial to preserve it."

Wagner tries hard to follow the example of Richard Dawkins as a science writer. Although the result is a surprisingly enjoyable read – especially given the dry matter at hand – it will never match its model. It lacks the civilised aggressiveness for defending a unique idea: the selfish gene against the muddled thought of group selection.

In contrast to Dawkins, Wagner clearly overstates his case, by claiming to "present the missing piece in Darwin's theory" on the cover. On the other hand, he clearly enables biologists to understand the paths that evolution takes to reach stunning diversity and adaptedness.

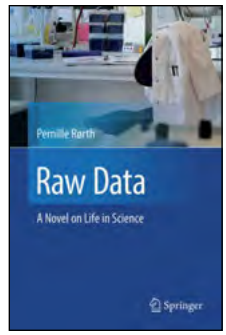
FLORIAN FISCH

Arrival of the Fittest: Solving Evolution's Greatest Puzzle. By Andreas Wagner. Penguin, 2014. 304 pages. €14.35 (paperback).

Book review: Envy-driven accusations and endangered careers

What Drives a Rising Star to Ruin It All?

Science is both utterly rewarding and frustrating. The temptation to cut corners is high. Follow Danish cell biologist and author, Pernille Rørth, into the exciting world of life sciences!



From its very first sentence *Raw Data* establishes a subtle sense of suspense. The reader dives directly into the microcosm of a top-level cancer research laboratory with its interactions between scientists, whose particular ambitions and intentions are only held together by a common aim: to advance their scientific field and to advance in it personally.

At first glance, the debut novel of the Danish cell biologist, Pernille Rørth, deals with the laboratory's everyday routine and gives insight into what makes a researcher 'tick'. The reader feels the enthusiasm with which aspiring young investigators plunge into their work, spending every free minute at the bench, but also suffer their self-doubt and disappointment when experiments fail. An immense pressure weighs on not-yet established researchers, a pressure built up by competition between different research groups, as well as colleagues within the same group, fostered by the pursuit of spectacular results and the need to publish in journals with the highest impact factors. Which scientist has never been haunted by the dictum 'publish or perish'? Publishing in a renowned journal – today more than ever before – paves the path to scientific success.

Written by a science insider

Rørth is just such a successful researcher who has, in her 25 years of active scientific life, published as a senior author in high impact journals such as *Nature*, *Science*, *PNAS*, *Cell*, *Current Biology* and *The EMBO Journal*, of which she has been Executive Editor for several years. Thanks to her work on how groups of specific cells are guided from one area in the organism to another, a knowledge that is indispensable for understanding the mechanisms of cancer metastasis, she is all too familiar with scientists' daily concerns. In 2009, Rørth had to retract a publication in *Current Biology* due to mistakes that she had uncovered herself.

It may be partly due to this unpleasant experience that she spices up her plot with a case of scientific misconduct. Karen, a postdoc struggling to set up her own innovative project, by chance reveals irregularities that suggest that her colleague Chloe has boosted her results just enough to get accepted for publication in *Nature*. Due to this publication, Chloe is poised to take the next step on the way to an independent research group.

It is one of the novel's assets that, next to a detailed and accurate description of the daily lab routine, with its clash of extremely ambitious and gifted individuals, neither the culprit nor the whistleblower – nor the apparently uninvolved principle investigator – are painted in black or white. With pronounced sensitivity, the novel elaborates the fatal circumstances that drive young researchers in the process of qualification for an independent research position to cross the boundary and violate ethical rules. The fact that success is almost solely measured in the number of publications, together with the power of the anonymous reviewers that literally dictate the missing results in the first author's laboratory notebook, creates pressure and temptation at once. The solution seems ever so easy. Conceding to this temptation is something, to which nobody seems to be immune.

A victim of circumstances?

A certain joint guilt is attributed to the way that journalists cover retractions and focus more on the unethical behaviour itself than on the reasons leading to it. Even if a paper is only retracted due to unintentional mistakes, as in the case of Rørth, a retraction remains an unpleasant experience. Even today, her retraction remains one of the first Google hits when you search for Rørth. Certainly nobody wants a retraction to be the first thing associated with their name, which may explain why mistakes and suspicions of scientific fraud often stay unreported. *Raw Data*'s plot is reminiscent of the Brazilian immunologist Thereza Imanishi-Kari's case, a scientist accused of data forgery in a *Cell* paper in 1986. All charges against her were dismissed ten years later and today she works as an associate professor at the private Tufts University in Massachusetts.

Keeping up the suspense

Other cases, however, lead to the free fall of highly respected scientists like the Korean stem cell researcher Hwang Woo-suk, a pioneer in his field and the "pride of Korea", before the falsification of his results about the production of human stem cells became obvious in 2006. The direction that Rørth's plot takes shall remain open at this point. Rørth manages to keep up the suspense: are we really reading about scientific misconduct or about a case of envy-driven false accusations? Step by step, mistrust creeps into the lab members' minds, poisons the atmosphere and events take their course until more than one career seems to be endangered.

With her book, Rørth intends to "give non-scientists a chance to see what life in science is like". It is difficult, however, to believe that a scientific illiterate would be sufficiently able or willing to get into the authentically-described scientific details in order to follow the plot. For those with a scientific background, on the other hand, this compelling story with its surprising end is definitively worth reading.

Larissa Tetsch

Raw Data. By Pernille Rørth. Springer, 2016. 195 Pages, €21 (softcover), €15 (eBook).

Career strategies for young European scientists

From Mutterland to Mutterland via Vaterland

Photo: Pixabay/Unsplash

Fascinated by Germany, Indian neuroscience graduate, Vasudharani Devanathan, packed up all her bags and started her PhD in Hamburg. After ten years in Europe, expanding not only her scientific skills, she's now back in India at a shiny new institute, bringing in all the know-how gained during her German adventure. Here's her story.

This is all about my move from India to Germany and back to India, hence the title. For the love of Deutsch and Deutschland, here and there a few German words will pop up in this article but don't worry, you will understand.

Just after a regular work day at AstraZeneca in Bangalore, I went to the nearest internet centre and had a surprise waiting for me in my mail box. I was very happy to receive an email from Melitta Schachner from the Zentrum für Molekulare Neurobiologie, Hamburg (ZMNH), in Germany.

Vasudha's Hamburg group. Here, without barbecue grill and Franzbrötchen.

My joy knew no bounds. The email conveyed the message that I will be called for a telephone interview and if selected, will be invited to start my research work at her lab with a three-month observation period. These three months, as I understood, are the personal interview period... probably the longest personal interview ever!

Curious about Germany

Like the majority of Indians, I was also prepared to go to the USA but 50% of my heart leaned towards Germany. For two reasons, first, my curiosity about Germany. Many of my friends and seniors told me that the US is a more friendly country but deep in my mind I was always convinced that Germany would be perfect for me. Second, I had been working for two years after my post graduation and was concerned that I had lost those two years. That's why I wanted to be in Germany where I knew it was possible to finish a PhD in 3.5 years – although, in the end, it took me about 4.5 years to complete!!

All went well during my interview and I landed at Hamburg airport on April 18th, 2003. The Italian postdoctoral scientist, with whom I was going to work, very kindly came to pick me up. All Italians are known to be friendly; she immediately started telling me in detail about Germany with its pros and cons for foreigners. The first thing she said was "Ordnung muss sein". It's not important how much you work, it is important how much you write in the methods book and organise your work!

What I understood initially and finally, after ten years of doing research in Deutschland (yes, after living there, I prefer to call it Deutschland rather than Germany) is that "the time taken to organise the work is inversely proportional to the time taken to perform experiments and directly proportional to success. A well-planned and well-documented experiment will require less or the optimal time to execute an experiment yielding a successful scientific solution".

Quiet Hamburg

It was Easter time in Hansestadt Hamburg (as it is addressed locally!!) and for a person from Chennai, Hamburg was totally empty. In Chennai, you bump into someone almost every minute and if you are "lucky", every 10 seconds! I was a bit nervous about how I would manage in a city, which is so silent and almost dead. The next morning, however, when I went to the city centre and

the Alster lake, my fears were dispersed. I saw life everywhere and such an incredibly accentuated spring, for the first time in my life! There were cherry blossoms in abundance.

My apartment was in the beautiful urban district of Eppendorf, just behind the Universitätsklinikum Eppendorf (UKE). My new workplace was situated just across the UKE campus, at the ZMNH. The Tuesday after Easter Monday, I joined the lab.

Our lab had about 30 PhD students and 15 post docs from all over the world. There were people from countries, I hadn't even heard of before joining this lab. For example, while having tea in the "Küche" on the third day, I met another PhD student, who told me she was from Georgia. "Atlanta, Georgia" I wondered? She jumped out of her chair and indignantly replied, "No Vasu, it is in Eastern Europe and it is older than that Georgia".

Refining more than one skill

Thus, not only my scientific skills developed but my geographical, cultural, ethnical knowledge developed tremendously, too. I opened up to new cultures and my tolerance for different kinds of people increased enormously. The first getaway with my lab mates was to the Hafengeburtstag (literal translation: harbour birthday). It is special and unique for Hamburg and celebrated with great joy there. Indeed, it was a memorable evening. We walked all along the harbour and on our way back, passed through the Reeperbahn, Hamburg's world-famous entertainment district.

Our lab life was simple: arrive at the lab at around 09.00 am, start experiments, have a cup of coffee with Franzbrötchen (Butterbrezel during my life in Tübingen). There were about ten of us having lunch together, which was always an exciting happening, with many varieties of food from different cuisines! Post-lunch meant going back to Arbeitszeit and then we had dinner in the lab, too. Normally, I would be back home around 10.00 or 11.00 pm. Amidst such scheduled lab times, we also tried to take small breaks for a jog along the Alster lake or in the Eppendorf park, or go for a quick swim at Holthusen-Bad or Kaifu-Bad.

Seminars and barbecues

We had some really stressful times, when experiments did not work for about two to three months. This usually meant a lot of brain storming, reading and discussion sessions! During my lab week, I also had two seminars. A smaller one, which only group members attended, and a big seminar with the entire lab. With such a huge lab, the big seminar sometimes looked like a mini symposium session at a conference! Barbecuing is a true German tradition and we were no exception. Every summer, there was a barbecue in the Eppendorf park, which I miss, even to this day!

All said and done, I completed my PhD with a Summa cum Laude defence and joined Bernd Nürnberg's lab for my post-doctoral training. I moved to Düsseldorf (my husband was working there), carrying a lot of memories from Hamburg. In the beginning, it was hard for me to accept the fact that I had to move away from the Alster lake, Hafen, Altona, Eppendorf and Hamburg's numerous cafeterias, pizzerias and pasta bars.

The next stop on Vasudha's German adventure: one of the most beautiful cities in Germany, Tübingen, with surprisingly little bureaucracy .

The lab in Düsseldorf was not as big as my PhD lab but had enough people to learn new things and interact. Düsseldorf is a beautiful city, too – very elegant, classy and the Altstadt walk is the best thing to do on a Saturday evening. Bernd Nürnberg is a gentle and kind person, and one can learn a great deal of things from him. Unfortunately, the lab soon moved to Tübingen and so did I.

The perfect town

After arriving at Tübingen, I quickly figured out that the move was not as unfortunate as I had thought. A city like Tübingen, with its romantic old architecture, the University spread throughout the town, hilly, picturesque landscape, really is how you would imagine your perfect scientific home town should be. Tübingen also hosts the only graveyard, where I could never feel sad, as in this place, many Nobel laureates and other scientific achievers had found their last resting place.

One big headache in any foreign country is the residence permit. And Germany is famous for its orderliness. But if you have the right documents, Germans are very helpful and quick. To put an end to the lengthy visa extension procedure, I applied for a Niederlassungserlaubnis, which was approved in the record time of about one hour. After submitting all the documents at the Bürgeramt, I came back to my lab and was surprised to receive a call, stating my application had already been accepted!

During our time in Germany, we also took the opportunity to travel to almost all European countries, exploring the Alps and the Black Forest. But the time came to move back to India, with a dream to do good science there as well. Looking back, it was the most precious time of my professional life – I built scientific collaborations and gained numerous friends from all over the world. When I think of Germany, I will always fondly remember the gentleness, professionalism, perfection and straightforwardness. Whoever says, Germans are not easy to make friends with, I will only say – it takes time, but they are friends forever!! Finally, 1st June 2012, it was Auf Wiedersehen, Deutschland!

Back in India, we moved from city to city due to my husband's job. I was on maternity leave, taking care of our son. After a three-year break, I started teaching at M.S. University in Baroda and also applied for research funding from the Department of Science and Technology India (DST), which was eventually granted. Since it was only a temporary position, I was on the look-out for something more appropriate for me. India has several institutes of scientific excellence and universities of academic excellence. But there are very few institutes that offer a research environment, at which the scientist can both perform research and participate in teaching, as is common in the German university system. I think

Careers in academia

Bringing together teaching, doing research, international expertise and day-care centre

this is an optimal model, as even an undergraduate student has the chance to be taught by an outstanding scientist, who excels in a specialised area.

Returning with a vision

To fill this gap between research and teaching, the Indian Institutes of Science Education and Research (IISERs) were established by the Government of India. IISERs offer an integrated BS-MS course in science. Any student, entering this course, will be required to study concepts in basic sciences (Biology, Chemistry, Physics and Maths) for four semesters, after which, they are allowed to specialise in the subject of their choice. Beyond this semester, several specialised courses (e.g., immunology, neuroscience, forensic science, astrophysics) are offered. IISERs host several visionary scientists, who are educated in highly reputable institutes and have returned to India with a vision to establish international scientific culture in India.

Only at the very last minute, I saw an advertisement from IISER Pune for faculty positions in the new IISER at Tirupati. Hurriedly, I sent in my application package, so as not to miss that opportunity. Fortunately, my application was shortlisted for an interview, which was to be held in Pune. Even for the interview, the people at the institute were kind and helpful. Telling them, I did not know where to leave my little son during my interview, they kindly offered me a place, for one day, in the day care centre within the campus. Only a few places in India have such a facility, perhaps very few places all over the world.

Kind reception

Arriving at the campus, I immediately felt welcome and was even interviewed first, so I wouldn't be away from my son for an unnecessary length of time. How thoughtful and friendly! After competitive selection, I was invited to join the new IISER team, which is mentored by the experienced IISER Pune team. Since I was going to move with my toddler, I was also offered best possible care for me to settle down in Tirupati. IISER in Tirupati (IISER T) is the sixth IISER Institute and started operation in August 2015. Mentored by IISER Pune, IISER T has completed its first year with its first batch of 48 students.

During the first semester, our mentors from Pune, Dean Bhas Bapat, Coordinator V.S Rao and A.A. Natu flew in and out of Tirupati and ensured that the scientific standards set in IISER Pune had been transferred to us. IISER Pune's director, KN Ganesh, takes personal care of our research needs and is also constantly monitoring the quality and standards of the research labs, currently being established in Tirupati.

The Biology lab, for instance, is going to be equipped with the latest instruments required for cutting edge research. Confocal and fluorescence microscopes, FACS, ultracentrifuges, BSL-2 cabinets for cell culture and a dedicated cell culture facility will soon arrive at the campus. Within one year, faculty and students have initiated several extra-curricular activities and clubs such as a science club, an eco club, biology club and maths club. Facilities at the institute include a sports room, a TV room, gym and whatnot, even a small place for our toddlers.

Know-how from across the globe

The best things about the Institute is that the faculty has brought with them expertise from different parts of the world and is also given full freedom to design courses, curriculum and any other activity in the Institute. To increase interaction between students and teacher, each of us has only five students, whom we closely mentor. In such an intimate surrounding, it's much easier to help the students study well and also take care of their emotional needs.

Apart from applying to various research funds and establishing biology labs, I also teach an undergraduate biology course, conduct exams and assess students. My position has allowed me to establish new collaborations with national and international institutes, which will hopefully advance my research in neurodegeneration and cell signalling. At my lab in IISER T, we will soon be taking PhD students. While India has opened further doors for me, Deutschland has not closed its doors. I still collaborate with my former bosses in Tübingen and Hamburg and also initiate MoUs [Memoranda of Understanding] between our institutes. Without exaggeration, I can confidently say that IISERs are to India what the Max Planck Institutes are to Germany.

Vasudharani Devanathan

If you want to share your story, write to editors@labtimes.org

Calendar

2016

27/6–28/6 London (UK)
Membrane Pores: From Structure and Assembly, to Medicine and Technology, *Info: https://royalsociety.org/events/2016/06/membrane-pores*

27/6–30/6 Örebro (SE)
Conference of the International Network of Environmental Forensics, *Info: www.rsc.org/events/detail/21538/the-international-network-of-environmental-forensics-inef-2016-conference*

28/6–30/6 London (UK)
Parasitic Infection 2016, *Info: http://lifescienceevents.com/parasitic2016*

28/6–1/7 Uppsala (SE)
European Chapter Meeting of the Tissue Engineering and Regenerative Medicine International Society (TERMIS-EU) 2016, *Info: www.termis.org/eu2016*

29/6–30/6 Dundee (UK)
Young Microbiologists Symposium on Microbe Signalling, Organisation and Pathogenesis, *Info: www.lifesci.dundee.ac.uk/other/yms*

29/6–1/7 Liverpool (UK)
Annual Meeting of the Bone Research Society (BRS), *Info: http://boneresearchsociety.org/meeting/liverpool2016*

30/6 London (UK)
London Chromatin Club, *Info: www.abcam.com/events/london-chromatin-club-june-2016*

30/6–1/7 Dublin (IE)
Host-Pathogen Interactions, *Info: http://poxasfairid2016.sciencesconf.org*

30/6–2/7 St. Petersburg (RU)
St. Petersburg International Symposium on Systems Biology and Bioinformatics (SBBI 2016), *Info: www.sbb16.spbstu.ru*

1–5/7 Bischenberg/Strasbourg (FR)
21st International Poxvirus, Asfarvirus and Iridovirus Conference, *Info: http://poxasfairid2016.sciencesconf.org*

2/7–6/7 Copenhagen (DK)
10th Forum of Neuroscience, Organised by the Federation of European Neuroscience Societies (FENS), *Info: http://forum2016.fens.org*

3/7–5/7 Toulouse (FR)
6th International Bacterial Wilt Symposium, *Info: https://colloque.inra.fr/ibws2016*

3/7–6/7 Krakow (PL)
European Congress on Biotechnology, *Info: http://ecb2016.com*

3/7–7/7 Uppsala (SE)
15th International Symposium on Amyloidosis, *Info: https://akkonferens.slu.se/isaamyloid2016*

3/7–8/7 Girona (ES)
Gordon Research Conference: Cell Death, *Info: www.grc.org/programs.aspx?id=12477*

3/7–8/7 Goettingen (DE)
22nd International Symposium on Plant Lipids, *Info: www.eurofedlipid.org/meetings/goettingen2016*

5/7 Warwick (UK)
Warwick Quantitative Biomedicine Programme Symposium: Medicine in the 4th Dimension, *Info: http://go.warwick.ac.uk/medicine4d*

5/7–6/7 Surrey (UK)
Translation UK 2016, *Info: www.biochemistry.org/Events/tabid/379/MeetingNo/SA183/view/Conference/Default.aspx*

5/7–7/7 San Michele all'Adige (IT)
1st International Symposium on Biotremology – Studying Vibrational Communication, *Info: http://eventi.fmach.it/biotremology2016*

5/7–7/7 Heidelberg (DE)
EMBL Conference: Lifelong Learning in the Biomedical Sciences, *Info: www.embl.de/training/events/2016/LLL16-01*

6/7–7/7 London (UK)
3rd Annual Allergies Conference, *Info: www.allergies-event.com/bps*

6/7–7/7 London (UK)
3rd Annual Peptides Conference, *Info: www.bps.ac.uk/news-events/future-scientific-meetings/2016/peptides*

6/7–8/7 Frankfurt/M. (DE)
Biochemistry 2016 – Shaping the Molecules of Life: Chemical Biology of Nucleic Acid and Protein Modifications, *Info: www.gdch.de/biochemistry2016*

6/7–9/7 Bordeaux (FR)
11th European Congress of Neuropathology (ECNP), *Info: http://ecnp2016.com*

Event and Recruitment Ads

Rates

Rates for ads with frames & logo

size (width x height in mm):	*basic rate b/w*
1/1 page (185 x 260 mm)	€ 1,950.-
1/2 page (90 x 260 or 185 x 130 mm):	€ 1,040.-
1/3 page (90 x 195 mm):	€ 830.-
1/4 page (90 x 130 mm):	€ 590.-
1/6 page (90 x 100 mm):	€ 480.-
1/8 page (90 x 65 mm):	€ 350.-

Other sizes on request

4 weeks online at our website included!

Colour surcharge: € 390.- to €1.100.-

Payment: All prices are without VAT.

Dates and Deadlines

Issue	*Publishing date*	*Ad closing*
4/2016	1 September	**22 July**
5/2016	6 October	**9 September**
6/2016	30 November	**3 November**

For further information please contact:
Lj-Verlag OHG,
Lab Times, Ulrich Sillmann
Merzhauser Str. 177,
79100 Freiburg, Germany
Phone +49-(0)761 292 58 85
Fax +49-(0)761 357 38
Email: jobs@labtimes.org

Please subscribe at **www.labtimes.org**

Lab Times is free of charge for non-profit institutions all over Europe. It is distributed to scientists and lab staff wherever they work: in universities, research units, private and public research institutes, industry, etc.
For companies and personal subscriptions (if you want us to send *Lab Times* to your home address) the subscription fee is €27.- per year (6 issues).

6/7–10/7 Strasbourg (FR)
EMBO Conference on Ribosome Structure and Function 2016, *Info: http://events.embo.org/16-ribo*

7/7–10/7 Cambridge (UK)
The Pathobiology of the Lysosome and Lysosomal Diseases Conference 2016, *Info: www.zingconferences.com/conferences*

8/7 London (UK)
Protein O-GlcNAcylation in Health and Disease, *Info: www.biochemistry.org/Events*

10/7–12/7 Sitges (ES)
Cell Symposia: Aging and Metabolism, *Info: www.cell-symposia-aging-metabolism.com*

10/7–14/7 Bath (UK)
International Narcotics Research Conference (INRC) 2016, *Info: www.inrcworld.org/2016/2016mtg.htm*

10/7–14/7 Hernstein (AT)
Conference on Colloidal, Macromolecular and Biological Gels: Formulation, Properties and Applications, *Info: www.engconf.org/conferences/chemical-engineering*

10/7–14/7 Vilnius (LT)
24th Biennial Meeting of the International Society for the Study of Behavioural Development, *Info: www.issbd2016.com/en*

10/7–15/7 Girona (ES)
Gordon Research Conference: Transglutaminases in Human Disease Processes, *Info: www.grc.org/programs.aspx?id=14564*

10/7–15/7 Lisbon (PT)
FASEB Conference on Post-Transcriptional Control of Gene Expression: mRNA Decay, *Info: www.faseb.org/src/micro/Site/mRNA/Home.aspx*

11/7–13/7 Reading (UK)
Leucine Rich Repeat Kinase 2: Ten Years Along the Road to Therapeutic Intervention, *Info: www.biochemistry.org/Events/tabid/379/MeetingNo/SA176/view/Conference/Default.aspx*

11/7–14/7 Lisbon (PT)
6th Conference on Physiology of Yeasts and Filamentous Fungi (PYFF6), *Info: http://groups.tecnico.ulisboa.pt/bsrg/pyff6*

11/7–22/7 Montpellier (FR)
Ion and Water Transport in Plants – MISTRAL Summer School 2016, *Info: http://www1.montpellier.inra.fr/ibip/mistral/Objective.html*

12/7–13/7 Cambridge (UK)
Extracellular Vesicles: Biology and Therapeutic Potential, *Info: http://selectbiosciences.com/EVE2016*

12/7–15/7 Istanbul (TR)
Annual Meeting 2016 of the European Federation for Pharmaceutical Sciences (EUFEPS), *Info: www.eufeps.org/node/99*

16/7–22/7 Girona (ES)
Gordon Research Seminar and Conference: Endothelial Cell Phenotypes in Health & Disease, *Info: www.grc.org/programs.aspx?id=13296*

17/7–22/7 Lisbon (PT)
FASEB Conference on Smooth Muscle, *Info: www.faseb.org/src/micro/Site/Muscle/Home.aspx*

18/7–19/7 Cambridge (UK)
Quantitative Stem Cell Biology: From Molecules to Models – 5th Cambridge Stem Cell Symposium, *Info: https://sci-events.eventhq.co.uk/quantitative-stem-cell-biology-from-molecules-to-models*

18/7–22/7 Liverpool (UK)
EMBO Conference 2016 on Viruses of Microbes, *Info: http://events.embo.org/16-virus-microbe*

21/7–22/7 Berlin (DE)
2nd International Conference on Innate Immunity and Immune System Diseases, *Info: http://innateimmunity.conferenceseries.com*

21/7–23/7 Berlin (DE)
5th European Immunology Conference, *Info: https://immunology.conferenceseries.com/europe*

21/7–25/7 Prague (CZ)
12th International Congress of Cell Biology: Exploring Cellular Structure and Function, *Info: http://iccb2016.org*

22/7–27/7 Manchester (UK)
EuroScience Open Forum Conference 2016, *Info: www.esof.eu*

24/7–26/7 Heidelberg (DE)
EMBL Conference: Microfluidics, *Info: www.embl.de/training/events*

25/7–29/7 Barcelona (ES)
1st Conference on Social Impact of Science, *Info: http://socialimpactscience.org/sis2016*

27/7 London (UK)
Conference on New Insights in Inflammation, *Info: www.bps.ac.uk/news-events/future-scientific-meetings*

27/7–5/8 Lisbon (PT)
The Networking Challenge – Annual Symposium of Biology Students in Europe (SymBioSE) 2016, *Info: http://symbiose2016.eu*

29/7–31/7 Dublin (IE)
Physiology 2016, *Info: www.bps.ac.uk/news-events*

30/7–5/8 Girona (ES)
Gordon Research Seminar and Conference: Flow & Transport in Permeable Media – Bridging the Gap Between Scales and Processes for Strongly Coupled Systems, *Info: www.grc.org/programs.aspx?id=11312*

31/7–4/8 Macclesfield (UK)
80th Harden Conference: Machines on Genes IV – Mechanisms of Actions of Large Macromolecular Machines on Genes Across Biological Scales, *Info: www.biochemistry.org/Events*

1/8–3/8 London (UK)
Molecular Biology of Archaea 5, *Info: www.microbiologysociety.org/conferences/focused-meetings.cfm*

1/8–3/8 Manchester (UK)
2nd International Conference on Parasitology, *Info: http://parasitology.conferenceseries.com*

4/8–5/8 Manchester (UK)
2nd World Congress on Biopolymers, *Info: http://biopolymers.conferenceseries.com*

4/8–5/8 Manchester (UK)
7th Annual Conference on Stem Cell & Regenerative Medicine, *Info: http://stemcell-regenerativemedicine.conferenceseries.com*

4/8–5/8 Oxford (UK)
Metalloproteinases and Their Inhibitors: Beginning, Past and Future, *Info: www.kennedy.ox.ac.uk/more-events*

4/8–6/8 Essex (UK)
Phenotyping for Photosynthesis and Productivity, *Info: www.essex.ac.uk/bs/conferences/phenotyping.aspx*

6/8–12/8 Girona (ES)
Gordon Research Seminar and Conference: Neurobiology of Brain Disorders, *Info: www.grc.org/programs.aspx?id=15136*

7/8–12/8 Maastricht (NL)
17th International Congress on Photosynthesis Research, *Info: www.ps2016.com*

8/8–10/8 Cambridge (UK)
Structural Aspects of Infectious Disease, *Info: www.biochemistry.org/Events*

8/8–11/8 Warwick (UK)
Limits of Perception: Advances in Bio-Imaging, *Info: www.physoc.org/bioimaging2016*

8/8–14/8 Spetses (GR)
EMBO Conference on Chromatin and the Environment, *Info: www.embo.org/events/conferences*

13/8–14/8 Girona (ES)
Gordon Research Seminar: Biomineralization – Exploring Mechanisms Behind Biomineral Formation and Function, *Info: www.grc.org/programs.aspx?id=15199*

14/8–19/8 Girona (ES)
Gordon Research Seminar & Conference: Biogenic Hydrocarbons & the Atmosphere, *Info: www.grc.org/programs.aspx?id=11012*

15/8–19/8 Moscow (RU)
21st European Meeting of the Paleopathology Association, *Info: www.21ppa2016.com*

16/8–20/8 Barsinghausen (DE)
12th International Adenovirus Meeting (IAM 2016), *Info: www.iam-2016.de*

16/8–24/8 Spetses (GR)
EMBL Conference on Molecular Mechanisms of Ageing and Regeneration: From Pluripotency to Senescence, *Info: http://events.embo.org/16-ageing*

18/8–20/8 London (UK)
2nd International Conference on Systems and Synthetic Biology, *Info: http://syntheticbiology.conferenceseries.com*

20/8–26/8 Girona (ES)
Gordon Research Seminar and Conference: Mechanisms of Epilepsy & Neuronal Synchronization – Aberrant Circuits to Impaired Function in Epilepsy, *Info: www.grc.org/programs.aspx?id=13762*

21/8–25/8 Leuven (BE)
28th Conference of European Comparative Endocrinologists, *Info: https://kuleuvencongres.be/CECE2016*

22/8–26/8 Montpellier (FR)
EMBO Conference: Nitrogen Nutrition of Plants (Nitrogen 2016), *Info: https://colloque.inra.fr/nitrogen2016*

24/8–27/8 Linz (AT)
20th European Congress on Alternatives to Animal Testing / 17th Annual Congress of EUSAAT (European Society for Alternatives to Animal Testing), *Info: www.eusaat-congress.eu*

25/8–28/8 Budapest (HU)
12th European Nitrogen Fixation Conference, *Info: http://enfc2016.hu*

26/8 Zürich (CH)
5th International Symposium on DNA-Encoded Chemical Libraries, *Info: www.biomacromolecules.ethz.ch/symposium.html*

27/8–30/8 Heidelberg (DE)
EMBL Conference: Transcription and Chromatin, *Info: www.embl.de/training/events*

28/8–1/9 Braga (PT)
32nd Symposium of the European Society of Nematologists, *Info: http://esn2016braga.com*

28/8–1/9 Cork (IE)
31st International Symposium on Chromatography (ISC 2016), *Info: www.isc2016.ie*

28/8–2/9 Innsbruck (AT)
20th IAC Cyanophyte/Cyanobacteria Research Symposium, *Info: www.uibk.ac.at/congress/iac-symposium-2016*

28/8–2/9 Lyon (FR)
16th European Microscopy Congress, *Info: http://emc2016.fr/en*

29/8–1/9 Chania, Crete (GR)
34th SMYTE (Small Meeting on Yeast Transport and Energetics), *Info: www.smyte.eu*

29/8–1/9 Zurich (CH)
20th EUCARPIA General Congress: Plant Breeding – The Art of Bringing Science to Life, *Info: www.eucarpia.org/general-congress.html*

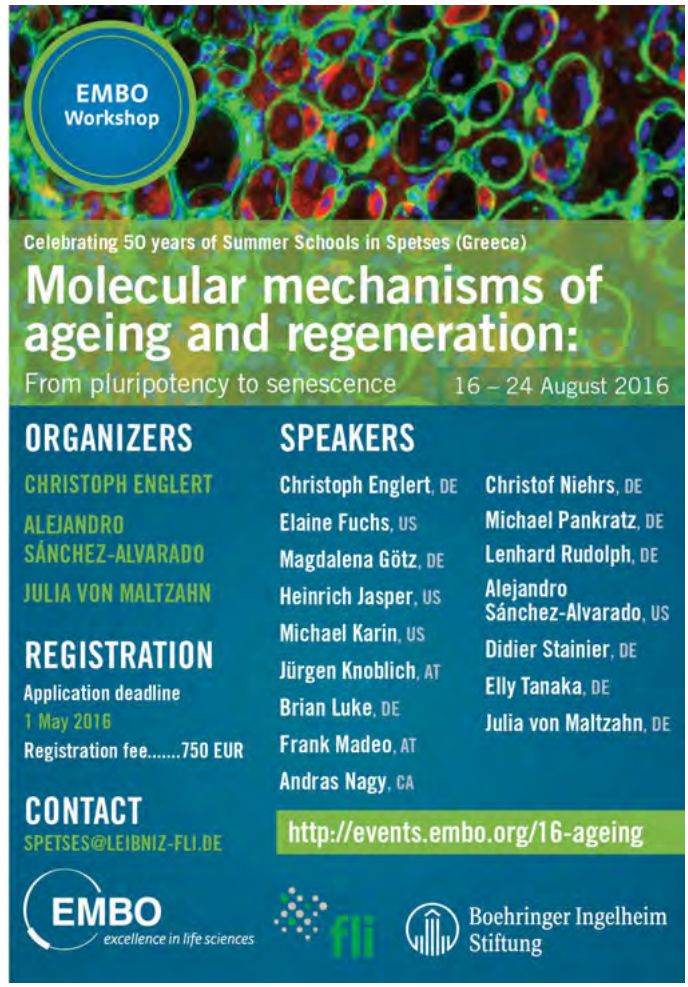
31/8–3/9 Heidelberg (DE)
EMBL Conference on Chemical Biology 2016, *Info: www.embl.de/training/events/2016/CHB16-01*

1/9–2/9 Dublin (IE)
Exploring the Microbe-Immune System Interface, *Info: www.microbiologysociety.org/events/focused-meetings.cfm*

3/9–7/9 Pontignano/Siena (IT)
EMBO Conference on Lymphocyte Antigen Receptor Signalling, *Info: http://events.embo.org*

3/9–7/9 The Hague (NL)
15th European Conference on Computational Biology (ECCB 2016), *Info: www.eccb2016.org*

3/9–8/9 Ephesus/Kusadasi (TR)
41st Congress of the Federation of European Biochemical Societies (FEBS), *Info: www.febs2016.org*



4/9–7/9 Ascona (CH)
2nd European Meeting on Phototransduction, *Info: www.uni-oldenburg.de/2nd-emp*

4/9–7/9 Barcelona (ES)
Unraveling Complexity: From Molecules to Ecosystems – 30th Congress of the New European Society for Comparative Physiology and Biochemistry, *Info: www.escpb.eu*

4/9–7/9 Seville (ES)
52nd European Congress of the European Societies of Toxicology, *Info: www.eurotox2016.com*

4/9–7/9 Winchester (UK)
81st Harden Conference: RNA and Disease, *Info: www.biochemistry.org/Events*

4/9–8/9 Bratislava (SK)
2nd International Plant Proteomics Organization World Congress, *Info: www.plant-phenotyping.org/inppo_congress_2016*

4/9–8/9 Les Diablerets (CH)
EUROPIC 2016 – Conference of the European Study Group of Picornaviruses, *Info: http://europic2016.org*

4/9–9/9 Manchester (UK)
20th International Pathogenic *Neisseria* Conference, *Info: www.ipnc2016.org*

5/9–7/9 Exeter (UK)
The Dynamic Fungus, *Info: www.microbiologysociety.org/events/focused-meetings.cfm*

5/9–8/9 Vienna (AT)
Dopamine 2016 – Dopamine Researcher Meeting of the Austrian Pharmacological Society (APHAR), *Info: www.dopamine2016.org*

6/9–7/9 Cardiff (UK)
GARNet 2016: Innovation in the Plant Sciences, *Info: http://garnet2016.weebly.com*

6/9–8/9 Web-based event
Moving Forward with Stem Cell Therapy, *Info: http://lifescience events.com/stemtherapy2016*

6/9–9/9 Oxford (UK)
8th Meeting of the European Society for *Chlamydia* Research, *Info: www.escr2016.co.uk*

7/9–8/9 Oxford (UK)
Phages 2016 Conference – Bacteriophage in Medicine, Food and Biotechnology, *Info: http://lpmhealthcare.com/phages-2016*

7/9–9/9 Cambridge (UK)
Wellcome Trust Conference on Exploring Human Host-Microbiome Interactions in Health and Disease, *Info: https://registration.hinxton.wellcome.ac.uk/Conferences.wt*

7/9–9/9 Sheffield (UK)
7th European Conference on Tetraspanins, *Info: www.biochemistry.org/Events*

7/9–10/9 Heidelberg (DE)
EMBO/EMBL Symposium on Actin in Action: From Molecules to Cellular Functions, *Info: www.embo-embl-symposia.org*

9/9–11/9 Estoril (PT)
2nd International Conference on New Concepts in B-Cell Malignancies, *Info: www.esh.org/conferences*

10/9–13/9 Mannheim (DE)
The EMBO Meeting 2016 – Advancing the Life Sciences, *Info: www.embo.org/events*

10/9–18/9 Dubrovnik (HR)
Microbial Diversity and Specialised Metabolites, *Info: www.jic.ac.uk/science/molmicro/Summerschool*

11/9–14/9 Dublin (IE)
44th Annual Meeting of the European Teratology Society, *Info: www.etsoc.com*

11/9–14/9 Liverpool (UK)
Pseudoenzymes 2016: From Signalling Mechanisms to Disease, *Info: www.biochemistry.org/Events*

11/9–16/9 Ascona (CH)
Liposomes, Exosomes & Virosomes – From Modeling Complex Membrane Processes to Medical Diagnostics & Drug Delivery: Biophysical Society Meeting, *Info: www.biophysics.org/2016switzerland*

12/9 Oxford (UK)
Emerging Viruses Symposium 2016, *Info: http://lpmhealthcare.com/emerging-viruses-2016*

12/9–14/9 Berlin (DE)
5th International Conference on Tissue Engineering and Regenerative Medicine, *Info: http://tissuescience-regenerativemedicine.conferenceseries.com*

12/9–14/9 Winchester (UK)
16th International Conference on Progress in Vaccination Against Cancer (PIVAC-16), *Info: www.eacr.org/pivac16*

12/9–16/9 Grenoble (FR)
4th International Soft Matter Conference *Info: www.ismc2016.org*

12/9–16/9 Roscoff (FR)
Protein Misfolding in Disease – Toxic Aggregation-prone Proteins in Aging and Age-related Diseases: From Structure to Pathology and Spreading (Conférences Jacques Monod), *Info: www.cnrs.fr/insb/cjm/2016/Buee_e.html*

13/9–14/9 Cambridge (UK)
2nd Annual Genome Engineering Congress, *Info: http://selectbiosciences.com/GE2016*

13/9–15/9 Oxford (UK)
Influenza 2016 Conference – One Influenza, One World, One Health, *Info: http://lpmhealthcare.com/influenza-2016*

14/9–16/9 Cambridge (UK)
Wellcome Trust Conference on Single Cell Genomics, *Info: https://registration.hinxton.wellcome.ac.uk/Conferences.wt*

14/9–16/9 Durham (UK)
Symposium on Microbial Protein Targets: Towards Understanding and Intervention, *Info: www.rsc.org/events/detail/20690*

14/9–17/9 Brno (CZ)
EMBO Conference: Wnt Meeting 2016, *Info: http://wnt2016.muni.cz*

14/9–17/9 Heidelberg (DE)
EMBL–Wellcome Trust Conference: Proteomics in Cell Biology and Disease Mechanisms, *Info: www.embl.de/training/events*

14/9–17/9 Kiel (DE)
Protease World in Health & Disease – 2nd International Symposium of the CRC877, *Info: www.uni-kiel.de/Biochemie/symposium2016*

15/9–16/9 Berlin (DE)
7th International Conference and Expo on Molecular and Cancer Biomarkers, *Info: http://molecular-cancer-biomarkers.conferenceseries.com*

16/9–20/9 Barcelona (ES)
17th International Conference on Systems Biology (ICSB 2016), *Info: http://icsb-conference.com*

17/9–20/9 Vienna (AT)
29th European College of Neuropsychopharmacology (ECNP) Congress, *Info: www.ecnp.eu/Congress2016/ECNP%20Congress.aspx*

17/9–21/9 Laura (Salerno) (IT)
EMBO Conference on the Molecular and Cellular Basis of Regeneration and Tissue Repair, *Info: http://events.embo.org/16-regeneration*

17/9–21/9 Portorož (PL)
15th International Symposium on Proteases, Inhibitors and Biological Control, *Info: www.eventgrids.com/events/2016_XVth_ISP_IBC*

18/9–20/9 Munich (DE)
4th Helmholtz-Nature Medicine Diabetes Conference, *Info: www.nature.com/natureconferences/hmgu2016*

19/9–22/9 Cambridge (UK)
Wellcome Trust Conference on Genome Informatics, *Info: https://registration.hinxton.wellcome.ac.uk/Conferences.wt*

19/9–23/9 Paris (FR)
EMBO Conference Tuberculosis 2016: Interdisciplinary Research on Tuberculosis and Pathogenic Mycobacteria, *Info: www.tuberculosis2016.org*

20/9–22/9 London (UK)
Rare Metabolic Disorders: Detection, Research, Management and Treatment, *Info: http://lifescience events.com/metabolism2016*

20/9–23/9 Basel (CH)
ILMAC 2016 – Trade Fair and Exhibition in Process and Laboratory Technology, *Info: www.ilmac.ch*

21/9–22/9 Oxford (UK)
Pharmacological Aspects of Microvascular Cell-Cell Signalling and CVS Disease, *Info: www.bps.ac.uk/news-events*

21/9–23/9 Bad Homburg (DE)
3rd European Platelet Network Conference – EUPLAN 2016, *Info: http://euplan.eu*

21/9–23/9 Oxford (UK)
Protein S-Palmitoylation: From Mechanism to Application, *Info: www.biochemistry.org/Events*

21/9–24/9 Barcelona (ES)
17th Biennial Meeting of the European Society for Immunodeficiencies (ESID), *Info: http://esid.org*

22/9–23/9 Paris (FR)
Final International Conference of the EUcelLEX Project (Cell-based ReGenerative Medicine: New Challenges for EU Legislation and Governance), *Info: www.eucellex.eu/final-international-conference*

22/9–23/9 Leuven (BE)
The Brain Mosaic: Cellular Heterogeneity in the CNS, *Info: www.vibconferences.be/events*

23/9 London (UK)
Beyond CpG Methylation: New Modifications in Eukaryotic DNA, *Info: www.biochemistry.org/Events*

23/9–24/9 Vienna (AT)
Platform f. Advanced Cellular Therapies Symposium: Designer Cells Go Clinic, *Info: www.pact.ac.at*

25/9–27/9 Heidelberg (DE)
EMBL–Wellcome Trust Conference: Big Data in Biology and Health, *Info: www.embl.de/training/events*

25/9–27/9 Seeon/Munich (DE)
Kloster Seeon Meeting on BACE Proteases in Health and Disease, *Info: www.bace-meeting.de*

25/9–28/9 Dubrovnik (HR)
Conference on Cell Fate Diversity in Aging, *Info: www.zingconferences.com/conferences*

25/9–29/9 Cologne (DE)
31st International Congress of the International Academy of Pathology and 28th Congress of the European Society of Pathology, *Info: www.esp-congress.org*

26/9–27/9 Newport Pagnell (UK)
Chemical Biology Approaches to Assessing and Modulating Mitochondria, *Info: https://royalsociety.org/events*

26/9–28/9 London (UK)
12th World Cancer Conference, *Info: http://cancer.global-summit.com/europe*

26/9–28/9 Montpellier (FR)
14th International Symposium on Rice Functional Genomics, *Info: http://isrfg2016.cirad.fr*

26/9–28/9 The Hague (NL)
4th International Conference on Responsible Use of Antibiotics in Animals, *Info: www.bastiaanse-communication.com/RUA16*

26/9–28/9 Ulm (DE)
Confocal Raman Imaging Symposium, *Info: www.witec.de/resources-and-education/symposium*

26/9–28/9 Valencia (ES)
2nd International Conference and Expo on Separation Techniques, *Info: http://separationtechniques.conferenceseries.com*

26/9–30/9 Rhodes (GR)
51st European Marine Biology Symposium, *Info: www.embs51.org*

27/9–29/9 London (UK)
Molecular Biology and Pathogenesis of Avian Viruses, *Info: www.microbiologysociety.org/conferences/focused-meetings.cfm*

27/9–29/9 London (UK)
Unlocking the Potential of Synthetic Biology to Enhance Human Health, *Info: http://lifescienceevents.com/syntheticbiology2016*

27/9–30/9 Athens (GR)
European Astrobiology Network Association Conference (EANA), *Info: www.astrobiology.gr/eana16*

28/9–1/10 Krk (HR)
Symposium on Power of Microbes in Industry and Environment, *Info: http://hmd-cms.hr/power2016*

28/9–30/9 Paris (FR)
International Conference on Nanomedicine and Nanobiotechnology (ICONAN 2016), *Info: http://premc.org/iconan2016*

28/9–3/10 Pula (Sardinia) (IT)
3rd Innovative Approaches for Identification of Antiviral Agents Summer School (IAAASS), *Info: http://people.unica.it/iaaass*

29/9–30/9 London (UK)
5th International Conference on Microbial Physiology & Genomics, *Info: http://microbialphysiology.conferenceseries.com*

2/10–7/10 Potsdam (DE)
EMBO Conference on Retinal Proteins, *Info: http://events.embo.org/16-retinal-proteins*

4/10–6/10 Bilbao (ES)
EMBO Conference on Translational Research in Cancer Cell Metabolism, *Info: http://events.embo.org*

4/10–7/10 Amsterdam (NL)
Cilia 2016, *Info: www.cilia2016.org*

5/10–7/10 Estoril (PT)
4th International Conference on Acute Myeloid Leukemia, *Info: www.esh.org/conferences*

5/10–8/10 Heidelberg (DE)
EMBO/EMBL Symposium on Complex Life of mRNA, *Info: www.embo-embl-symposia.org/symposia/2016/EES16-08*

6/10–8/10 Luxembourg (LU)
Annual Meeting of the Genetic Epidemiology of Parkinson's Disease Consortium and 3rd International Parkinson's Disease Symposium, *Info: https://parkinson2016.uni.lu*

10/10–12/10 Amsterdam (NL)
5th Beneficial Microbes Conference – Beneficial Impact of Pre- & Probiotics on Human and Animal Health, *Info: www.bastiaanse-communication.com/BMC2016*

10–12/10 Ebsdorfergrund (DE)
2nd Discussion Meeting Microbial Cell Biology, *Info: www.synmikro.com/de/startseite/86-termine*

10/10–14/10 Roscoff (FR)
Evolutionary Genomics & Systems Biology: Bringing Together Theoretical & Experimental Approaches, *Info: www.cnrs.fr/insb/cjm/2016/Vekemans_e.html*

11/10–12/10 Barcelona (ES)
Immune Profiling World Congress, *Info: http://www.terrapinn.com/events/life-science-and-health*

11/10–14/10 Groningen (NL)
5th International Conference on Novel Enzymes, *Info: www.rug.nl/research/gbb/education/novelenzymes*

12/10–15/10 Heidelberg (DE)
EMBO/EMBL Symposium on Organoids: Modelling Organ Development and Disease in 3D Culture, *Info: www.embo-embl-symposia.org*

Lab Tales

Number One

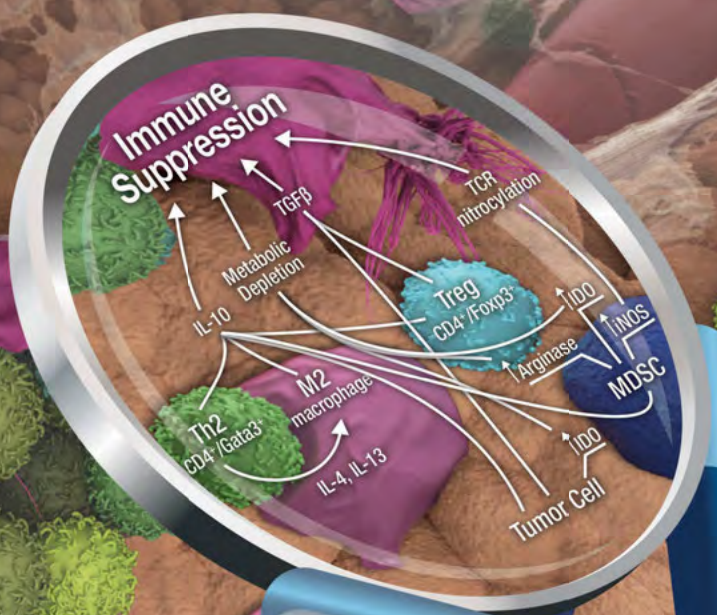

Labtimes online
http://www.labtimes.org/
CELL SIGNALING TECHNOLOGY
CST Guide:
Featuring Workflows
for PTM Research
SERVA
Electrophoresis
20% OFF
June
Lab Times
online
Search this Site
Rankings...
Products...
Methods...
Congresses...
Analysis...
EU Funding...
Careers...
Lab Tricks...
Cartoons...
Book Reviews...
ePaper...
Start
Funding
Methods
Products
Back Issues
Congresses
About Us
Subscribe
Current Issue
Rankings
Lab Tricks
Book Reviews
Cartoons
Careers
Media Kit
Contact
Online Editorials
Honey, I Shrunk the Organs
(June 8th, 2016) This year's Körber European Science Prize goes to Dutch professor of molecular genetics, Hans Clevers, for developing a procedure to grow miniature guts and livers from adult stem cells.
more...
for online viewing.
Table of Content
ePaper
PDF-Download
EM-Tippspiel
Tolle Tagespreise
und Hauptgewinne!
Wou
find out
conductin
more...
Comput
expected to fr
manuscripts fro
more...
The Dark Si
(June 1st, 2016)
down; scientists lo
of research articles
to the research co
to come out of hiding?
more...
Click here for previous editorials.
study Alzheimer's
disease, the
discipline's hottest topic... more
Laboratory Tales
The Dark Side of Research Antibodies
Research Antibodies in a Nutshell

CELL SIGNALING TECHNOLOGY

Deciphering Cancer

Tools to Interpret the Tumor-Immune Cell Crosstalk

PD-1
IDO
PD-L1
LAG3
PD-L2
ICOS
TIM-3
CD3
Arginase-1
CD8
VISTA



Visit our website to request our **Tumor Immunology Poster**

www.cellsignal.com/immunetumor

For Research Use Only. Not For Use In Diagnostic Procedures.
© 2016 Cell Signaling Technology, Inc. Cell Signaling Technology and CST are trademarks of Cell Signaling Technology, Inc.

16PADIMMU0017ENG

Cell Signaling TECHNOLOGY®