

NECTAR 25th Annual Meeting
Lund, Sweden, December 9th-11th, 2015 Booklet

9th – 11th DECEMBER 2015

NECTAR

25TH ANNUAL MEETING
LUND, SWEDEN



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A message from NECTAR President, Dr. Eilís Dowd

On behalf of Prof. Malin Parmar, Prof. Roger Barker, Prof. Anders Björklund and Prof. Cecilia Lundberg, the Local Organising Committee for NECTAR 2015, and the NECTAR Board, it gives me great pleasure to welcome you all to Lund for this very special NECTAR conference.

The founding NECTAR meeting was held in Le Vesinet, France in 1991, and this year's conference is the 25th gathering of our network. To mark this landmark occasion, Malin and her colleagues have put together a special 3-day programme with NECTAR's now trademark blend of talks from scientists and clinicians, as well as insights from patients and ethicists. And, for those of us who weren't at Le Vesinet in 1991 (the vast majority of this audience!), we are also going to be treated, by Anders and Steve, to their personal perspectives on the formation and evolution of NECTAR at the conference dinner.

We have come a long way since Le Visinet – in 1991, could anyone have conceived of turning a skin cell into a functional neuron for neuronal replacement, or could anyone have envisaged the in situ reprogramming of glial cells into functionally integrated neurons? The field of neural protection, repair and regeneration has come an extraordinarily long way in the intervening years, and NECTAR scientists have been, and continue to be, at the forefront of many of these remarkable discoveries. It is an exciting time to be in this field, and protective, reparative and/or regenerative therapies must surely be on the horizon for patients.

Also, we just wanted to take this opportunity, on behalf of the NECTAR Board and the wider NECTAR community, to express our thanks to our immediate past president, Dr. Emma Lane. Emma was an transformative NECTAR President, and her legacy will continue for many years to come. In particular, Emma's establishment of the NECTAR website has provided a centralised online resource which has greatly facilitated the organisation of the annual conferences, and she can also be thanked for bringing NECTAR into the social media domain through establishment of the NECTAR Facebook and Twitter pages. Thanks Emma!

So don't forget to tweet any comments, observations or photos you may have throughout this year's conference: @NECTAR_EU, #NECTAR2015.

Have a great conference and enjoy beautiful, Christmassy Lund!

With very best wishes,

Eilís Dowd

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Disclaimer

The local and scientific organizing committees accept no liability for injuries and losses of whatever nature incurred by participants and/or accompanying persons, nor loss of, or damage to, their luggage and/or personal belongings.

2015 NECTAR programme

Day 1 – Wednesday, December 9th, 2015

Talk	Speaker	Time
Welcome: Eilis Dowd		14.00
Session 1: Stem cell therapy for PD		
<i>Chair: Anders Björklund</i>		
Fetal and stem cell grafts for Parkinson's disease: Successes and Cautions	Jeffrey Kordower	14.10-14.40
Transplant-derived dopamine re-innervation of putamen after 24 years in Parkinson disease	Jia-Yi Li	14.40-15.10
Coffee 15.10 -15.40		
Are stem cell therapies ever likely to be a competitive treatment option for PD?	Roger Barker	15.40-16.10
Challenges towards stem cell therapy for Parkinson's disease	Jun Takahashi	16.10-16.40
Datablitz – Session 1		
<i>Chair: Simon Stott</i>	8 speakers (4 min+2 min)	16.45-17.40

18.30 Social activity for pre-registered participants

19.00 Dinner with invited guests: Gattostretto, Kattesund 6A

Day 2 – Thursday, December 10th, 2015

Talk	Speaker	Time
Session 2: Progress in cellular re-programming		
<i>Chair: Johan Jakobsson</i>		
Making new neurons via cellular reprogramming	Malin Parmar	9.00-9.30
iPS cells and Parkinson's disease: from disease modeling to drug discovery	Jared Sternecker	9.30-10.00
Datablitz – Session 2	<i>8 speakers</i>	10.00-10.50
<i>Chair: Aideen Sullivan</i>	<i>(4 min+2 min)</i>	
Coffee 10.50-11.20		
Transcription factor-mediated neuronal forward programming of human stem cells	Volker Busskamp	11.20-11.50
Transplanted embryonic neurons integrate into adult neocortical circuits	Sofia Grade	11.50-12.20
Lunch 12.20-13.10		
Datablitz – Session 3	<i>8 speakers</i>	13.10-14.00
<i>Chair: Mariah Lelos</i>	<i>(4 min+2 min)</i>	
Round Table Ethics session lead by Göran Hermerén with invited guests Inez de Beaufort, Jonathan Kimmelman		14.00-15.30
Coffee 15.30-16.00		
Session 3: Neurogenesis and neuronal remodeling		
<i>Chair: Malin Parmar</i>		
CRISPR/Cas9 editing of hPSC for modeling disease and human neural development	Meng Li	16.00-16.30
Generating and regenerating the cerebral cortex: from neuron subtype development and diversity to circuit implementation at the growth cone	Jeffrey Macklis	16.30-17.00
Progenitor cell-based treatment and modeling of glial disorders	Steven Goldman	17.00-17.30
Datablitz – Session 4	<i>5 speakers</i>	17.30-18.00
<i>Chair: Rosemary Fricker</i>	<i>(4 min+2 min)</i>	
19.00 Conference dinner at Pelarsalen, main University building		

Day 3 – Friday, December 11th, 2015

8.30 – 8.50 *St. Lucia's celebration with a candlelit choir*

Talk	Speaker	Time
Session 4: A patient-centered session		
<i>Chair: Roger Barker</i>		
Patient session including participation of a patient	Håkan Widner	9.00-9.30
How to deal with the unknown risks	Nils-Eric Sahlin	9.30-10.00
Doctors and Patients – Knowing Me, Knowing You...	Tom Isaacs	10.00-10.30
Coffee 10.30-11.00		
Realistic expectations?: Patients and public representations of (unproven) stem cells	Timothy Caulfield	11.00-11.30
Datablitz – Session 5	<i>6 speakers</i>	11.30-12.10
<i>Chair: Daniella Rylander Ottosson</i>	<i>(4 min+2 min)</i>	
Lunch 12.10-13.10		
Session 5: Cell and gene therapy for degenerative diseases		
<i>Chair: Christian Winkler</i>		
Repairing the diseased retina with stem cells	Rachael Pearson	13.10-13.40
Regenerative medicine, stroke	Zaal Kokaia	13.40-14.10
Working towards the production of authentic human striatal neurons from stem cells	Elena Cattaneo	14.10-14.40
Datablitz – Session 6	<i>7 speakers</i>	14.40-15.30
<i>Chair: Marie Jönsson</i>	<i>(4 min+2 min)</i>	
Close of the meeting	Roger BARKER Malin PARMAR	15.30

NECTAR – 25 YEARS IN PERSPECTIVE

Now when Nectar holds its 25th meeting in Lund it is timely to look back and remember how it all started. It was the launch of the first EU-sponsored research program in 1990, called BIOMED 1, that opened up for European funding of joint initiatives in the biomedical field. In this call researchers from member states across the union could request money for interactions and exchange, not for actual research. A group of us, including me, Steve Dunnett, Olle Lindvall, Patrik Brundin, Wolfgang Oertel, Andreas Kupsch and Gilles Defer, met in 1990 in Wolfgang's office in Munich to plan for an application to this call. It received very positive reviews and was funded for four years.

In this grant proposal we included in the budget the costs for a joint network meeting to be held annually at different sites in Europe. Although Marc Peschanski did not attend the planning meeting in Munich he volunteered to host the first annual meeting, which was held in Le Vesinet outside Paris on 10-12th May, 1991. It was at this meeting Steve proposed to give our network a name and came up with the acronym NECTAR for Network for European CNS Transplantation and Restoration, and Niall Quinn in London took on the job to submit a second EU application coined Nest-HD, which was focused on cell based therapy for Huntington's disease.

These two programs helped to finance the first six NECTAR meetings hosted by Vincenzo Silani in Milano in 1992, by Jens Zimmer at Sandbjerg Manor in Denmark, also in 1992, by Marc Livivier in Brussels in 1993, by Gerard Boer in Amsterdam in 1994, and by Harry Steinbusch in Maastricht in 1995. At that point it had become clear that the NECTAR meetings had become an essential and much valued part of the European brain repair effort, and although EU funding had expired everyone was keen to keep the meetings going. But the conditions were now different: the local organizers had to volunteer to generate funds for the local expenses, and the participants had to cover their own travel costs.

During the early years NECTAR undertook two tasks that turned out to be very important. The first one was the development of ethical guidelines for the use of human fetal tissue for transplantation and therapy. Gerard Boer in Amsterdam took the lead in this work.

Following discussions within NECTAR, and after external review and consultations, these guidelines were finally published in Journal of Neurology in 1994. The second task was the development of criteria for the clinical assessment of transplanted PD patients. Together with Bill Langston in San José, Håkan Widner took the lead in this work, resulting in the so-called CAPIT protocol that was published in Movement Disorders in 1992, and followed a few years later by a modified version, called CAPSIT-PD3.

These two NECTAR products have had considerable impact. The ethical guidelines have been important not only as a self-regulating document within the research community, but it has also inspired and influenced the discussions within political circles and legal bodies across Europe. Thanks to this initiative the use of fetal tissue in translational research

became at an early stage well regulated in many European countries, giving Europe a clear advantage in this field of research. The CAPIT (and CAPSIT-PD) clinical assessment protocols have provided a joint basis for long-term evaluation of PD patients, not only in transplantation trials but it has also been widely used for the assessment of PD patients undergoing other types of surgical interventions, such as deep-brain stimulation⁴. NECTAR has over the years been successful in adapting to the rapid developments that have taken place in the brain repair field, always with the focus on efforts aimed at the development of therapeutic interventions in patients with PD, HD and other neurodegenerative diseases. The NECTAR meetings have been, and continue to provide, a unique forum for interaction between clinicians and basic researchers in this dynamic field. Many successful collaborations have grown out of discussions held at NECTAR meetings, and most importantly perhaps, they have helped to foster the uniquely friendly and collaborative spirit that is such an attractive feature of our field. As we are entering an exciting new era in the application of stem cell research for restorative therapies I am sure that the NECTAR meetings will continue to play a central role.

Lund in December 2015
Anders Björklund

- 1 Boer GJ. Ethical guidelines for the use of human embryonic or fetal tissue for experimental and clinical neurotransplantation and research. *Journal of Neurology* 242:1-13 (1994).
- 2 Langston JW, Widner H, Goetz CG, Brooks D, Fahn S, Freeman T, Watts R. Core assessment program for intracerebral transplantations (CAPIT). *Mov Disord* 7, 2-13 (1992).
- 3 Defer GL, Widner H, Marié RM, Rémy P, Levivier M. Core assessment program for surgical interventional therapies in Parkinson's disease (CAPSIT-PD). *Mov Disord*. 14(4):572-84 (1999).
- 4 A search on Google Scholar using "CAPIT protocol", "CAPSIT protocol" and "CAPSIT-PD protocol" gives 223 hits.

ORGANISING COMMITTEES

International Organizing Committee (NECTAR board)

Eilis Dowd (President)

Emma Lane

Lachlan Thompson

Roger Barker

Paola Piccini

Gesine Paul

Malin Parmar

Tobias Piroth

Rosemary Fricker

Christine Kaindlstorfer

Local Organizing Committee

Malin Parmar

Roger Barker

Anders Björklund

Cecilia Lundberg

Paulina Pettersson

BIOGRAPHY OF SPEAKERS

Roger	BARKER	Cambridge, UK
Volker	BUSSKAMP	Dresden, Germany
Elena	CATTANEO	Milan, Italy
Timothy	CAULFIELD	Alberta, Canada
Inez	DE BEAUFORT	Rotterdam, the Netherlands
Steven	GOLDMAN	Rochester, NY, USA
Sofia	GRADE	München, Germany
Göran	HERMERÉN	Lund, Sweden
Tom	ISAACS	London, UK
Jonathan	KIMMELMAN	Montreal, Canada
Zaal	KOKAIA	Lund, Sweden
Jeffrey	KORDOWER	Chicago, IL, USA
Jia-Yi	LI	Lund, Sweden
Meng	LI	Cardiff, Wales, UK
Jeffrey	MACKLIS	Cambridge, MA, USA
Malin	PARMAR	Lund, Sweden
Rachael	PEARSON	London, UK
Nils-Eric	SAHLIN	Lund, Sweden
Jared	STERNECKERT	Dresden, Germany
Jun	TAKAHASHI	Kyoto, Japan
Håkan	WIDNER	Lund, Sweden

Roger Barker

Roger Barker is the Professor of Clinical Neuroscience, Honorary Consultant in Neurology at the University of Cambridge at Addenbrooke's Hospital and Guest Professor at Lund University. He trained at Oxford and London and has been in his current position for over ten years having completed an MRC Clinician Scientist Fellowship just prior to this. His main interests are in the neurodegenerative disorders of the nervous system in particular Parkinson's disease and Huntington's disease. He combines basic research looking at cell therapies to treat these conditions with clinically based work on defining the natural history and heterogeneity of both Huntington's disease and Parkinson's disease and is the co-ordinator of the EU FP7 TRANSEURO project looking at fetal cell grafting in patients with early PD.

Volker Busskamp

Dr. Volker Busskamp is a research group leader and a "Freigeist" fellow at the Center for Regenerative Therapies of the Technical University Dresden (CRTD), Germany. His research aims to drive human induced pluripotent stem (iPS) cells to neurons for disease modeling with a focus on entire neuronal circuit assembly to obtain sophisticated human test beds.

His group specifically develops and applies protocols to generate neuronal cell types of therapeutic interest from human iPS cells that will also be subjected to cell replacement therapies.

Volker Busskamp graduated in Biotechnology at the Technical University Braunschweig in Germany in 2006 and performed his postgraduate studies in Biology in Geneva, Switzerland in 2007. In 2010, he finished his PhD work in the laboratory of Botond Roska at the Friedrich Miescher Institute of Biomedical Research in Basel, Switzerland, on optogenetic repair of mouse and human photoreceptors. In addition, he studied the function of non-coding miRNAs in photoreceptors in health and disease. In 2011, Volker Busskamp joined George Church's laboratory at the Harvard Medical School in Boston, USA for his postdoctoral training. He focused on revealing the biological rules of human iPS neurogenesis applying systems biology. In 2014, he started his independent research position at the CRTD funded by the Volkswagen Foundation "Freigeist" program.

Elena Cattaneo

Elena Cattaneo is director of the "Laboratory of stem cell biology and pharmacology of neurodegenerative diseases" at the Department of Biosciences (www.cattaneolab.it) as well as co-founder and first appointed Director of UniStem (www.unistem.it), the Centre for Stem Cell Research of the University of Milano.

The main research theme of her lab is the molecular pathophysiology of Huntington's Disease (HD). The lab's ultimate goal is to identify cells, molecules and pathways that are suitable for therapeutic intervention and new reagents for drug screening in Huntington's Disease.

Over the years, the Cattaneo's lab was funded by (among others) the Huntington's Disease Society of America (USA), the Hereditary Disease Foundation (USA), the CHDI Foundation (USA), the European Union (through EuroStemCell, ESTOOLS, NeuroNE, STEM-HD, Stemstroke, Neuromics, Neurostemcell, Neurostemcellrepair projects), the Ministry of Research and University (Italy), Fondazione Cariplo (Italy), Telethon (Italy). E. Cattaneo has recently acted as coordinator of the EU project Neurostemcell (2008-2013). She is currently coordinating a new EU consortium – Neurostemcellrepair, 2013-2017 - as well as an Italian network on stem cells for Huntington's disease funded by the Ministry of Research and University (2013-2016).

On August 30th 2013, President of Italian Republic, Giorgio Napolitano, appointed her Life Senator on account of her scientific and social merit. Following the appointment, Elena Cattaneo has continued to lead the lab, within transcontinental, European and national research consortia. The aim remains to find breakthroughs that may accelerate a cure for Huntington's disease. She maintains her teaching position – with the exception of the formal entitlement of courses – and continues to lead the Unistem dissemination centre, working with scientific colleagues and Italian intellectuals to support and enhance science.

Timothy Caulfield

Timothy Caulfield is a Canada Research Chair in Health Law and Policy, a Professor in the Faculty of Law and the School of Public Health at the University of Alberta and Research

Director of the Health Law Institute at the University of Alberta. Over the past several years he has been involved in a variety of interdisciplinary research endeavours that have allowed him to publish over 300 academic articles. He is a Fellow of the Trudeau Foundation and the Principal Investigator for a number of large interdisciplinary projects that explore the ethical, legal and health policy issues associated with a range of topics, including stem cell research, genetics, patient safety, the prevention of chronic disease, obesity policy, the commercialization of research, complementary and alternative medicine and access to health care. Professor Caulfield is and has been involved with a number of national and international policy and research ethics committees. He has won numerous academic awards and is a Fellow of the Royal Society of Canada and the Canadian Academy of Health Sciences. He writes frequently for the popular press and is the author of two recent national bestsellers: *The Cure for Everything: Untangling the Twisted Messages about Health, Fitness and Happiness* (Penguin 2012) and *Is Gwyneth Paltrow Wrong About Everything?: When Celebrity Culture and Science Clash* (Penguin 2015).

Inez de Beaufort

Inez de Beaufort is a Professor of Medical Ethics at the Erasmus Medical Center in Rotterdam, the Netherlands. She studied theology at the University of Utrecht, Netherlands, where she specialized in ethics, wrote her PH on human research in Groningen and worked as a senior researcher at the Institute of Bioethics at Maastricht. She has published about, among others, reproductive ethics, beauty and ethics, fiction and ethics, decisions about the end of life, personal responsibility for health and obesity. She is a member of (among others) the European Group on Ethics in Science and New Technologies, honorary member of the Dutch Health Council and member of a Euthanasia Review Committee. She has organized the 11th conference of the International Association of Bioethics in 2012. She is a member of the Standing Government Committee for Reassessment of Parenthood. She has been involved in different EU funded projects as coordinator or as member of ethical advisory board.

Steven Goldman

Dr. Steven A. Goldman is the URM C Distinguished Professor of Neuroscience and Neurology at the University of Rochester Medical Center, and Co-Director of its Center for Translational Neuromedicine, in which he also holds the Dean Zutes Chair in Biology of the Aging Brain. He holds a concurrent appointment as Professor of Neuroscience and Neurology at the University of Copenhagen, where he is Co-Director of its Center for Basic and Translational Neuroscience, and serves as a consultant at Rigshospitalet, the Copenhagen University Hospital. Goldman moved to Rochester in 2003 from the Weill Medical College of Cornell University, where he was the Nathan Cummings Professor of Neurology. A summa cum laude graduate of the University of Pennsylvania, he obtained his PhD in neurobiology at the Rockefeller University in 1983, and his MD from Cornell in 1984. Goldman interned in Medicine and did his residency in Neurology at New York Hospital-Cornell and the Memorial Sloan-Kettering Cancer Center, before joining the Cornell faculty. Goldman is interested in

cell genesis and regeneration in the adult brain, with a focus on the use of stem and progenitor cells in treating demyelinating and neurodegenerative diseases. His lab focuses on the use of patient-derived stem and progenitor cells for the modeling of the neurodegenerative and neuropsychiatric disorders, as well as for the cell-based treatment of the pediatric leukodystrophies and multiple sclerosis. Goldman is a recipient of the Jacob Javits Neuroscience Investigator Award of the NIH, and was awarded the 2014 Novo Nordisk Foundation Laureate Award with Maiken Nedergaard. Among other honors, he has been elected to the Association of American Physicians, the American Society for Clinical Investigation, and Academia Europaea. Goldman remains active clinically, with subspecialty interests in neuro-oncology and myelin disease; he is an emeritus chairman of Rochester's Department of Neurology, founding director of its neuro-oncology program, and a former member of the FDA's Cell, Tissue and Gene Therapy Advisory Committee.

Sofia Grade

Sofia Grade, Ph.D. obtained her Ph.D. degree from the University of Coimbra, Portugal, in 2012. Her early interests concerned approaches for cell replacement in the injured brain, namely induction of oligodendrogenesis from postnatal neural stem cells to counteract demyelinating injuries, a work that she developed at the University of Coimbra, Portugal, and also the spontaneous recruitment of neurons from neurogenic niches to a stroke area in the adult brain, a project she developed at the Laval University, Québec, Canada. Since 2012 she is a postdoctoral fellow in Magdalena Götz's group at the Ludwig-Maximilians University and Helmholtz-Zentrum Munich, Germany. Dr. Grade's current research focuses on neuronal transplantation strategies in areas of pathological neuronal cell loss and assessment of the synaptic integration of those transplanted neurons in the host injured brain.

Göran Hermerén

Göran Hermerén, Fil. Dr, is professor emeritus of Medical ethics in the Faculty of medicine, Lund University, Sweden, previously professor of philosophy. From 2002 to 2011 he served as president of the European Group on Ethics in Science and New Technologies, Brussels, and is presently a member of the National Council on Medical Ethics, Stockholm, as well as chair of the permanent working group for science and ethics of ALLEA (All European Academies). Recent publications include papers on the principle of proportionality, fraud and misconduct in science, withdrawal from biobank research as well as books and anthologies on topics such as the goals of medicine, translational stem cell research, and trust and confidence in scientific research. He has been and is involved in many EU-funded projects, such as EuroStemCell, NeuroStemCell, ESTOOLS, RETHRIM and others, working on ethical aspects of stem cell research.

Tom Isaacs

Tom Isaacs is the President and a Co-founder of The Cure Parkinson's Trust, London, UK. Diagnosed with Parkinson's at the age of 27, Tom has done everything he can to raise funds, heighten awareness and find a cure for the condition. Having completed his successful 1,250

mile sponsored walk in 1999, Tom left his job as Director of a London property company in April 2002 to undertake his Coastin' challenge. By April 2003, Tom had walked 4,500 miles around the British coastline, climbed the highest mountains in England, Scotland and Wales and ran the London Marathon, raising over £350,000 for Parkinson's research. The Cure Parkinson's Trust has gone on to direct over £5 million into ground breaking Parkinson's research. Tom was a Board Member of the European Parkinson's Disease Association from 2005-2010. He is a patient representative for DeNDRoN and the Program Committee for the World Parkinson Congress 2016. He was also a leading contributor to the SENSE-PARK, an EU funded project to establish more personalised, objective measuring devices for people with Parkinson's and those who treat them.

Tom Isaacs is also co-founder of Parkinson's Movement, launched in 2011 to rally the voices of people with Parkinson's and ensure those living with the condition have influence, not just on their own lives but on the entire Parkinson's agenda – particularly in terms of building partnerships between those undertaking research and those participating in it.

Jonathan Kimmelman

Jonathan Kimmelman is Associate Professor in the Biomedical Ethics Unit / Social Studies of Medicine. He has cross appointments in Experimental Medicine, Epidemiology, Biostatistics and Occupational Health, and Human Genetics. Jonathan holds a PhD in Molecular Biophysics and Biochemistry from Yale University, and joined McGill in 2005. His research revolves around the ethical, social and policy dimensions of translational research. In 2006, he received the Institute of Genetics Maud Menten New Investigator Prize and currently holds a CIHR New Investigator Award. Jonathan chaired the ethics committee of the American Society of Gene and Cell Therapy, 2008-2010, and chairs the ethics committee of the International Society of Stem Cell Research. He also served on the CIHR Stem Cell Oversight Committee, is a current member of the Gene and Cell Therapy DSMB of U.S. National Heart, Lung, and Blood Institute, and is a member of the U.S. Institute of Medicine Committee on Ethics Principles and Guidelines for Long Duration and Exploration Spaceflights. His book, *Gene Transfer and the Ethics of First-in-Human Trials: Lost in Translation*, was published by Cambridge University Press.

Zaal Kokaia

Zaal Kokaia is a Professor of Experimental Medical Research at Lund University. He is currently the Director of Lund Stem Cell Center and head of Laboratory of Stem Cells and Restorative Neurology. He coordinates Strategic Research Area in Stem Cells and Regenerative Medicine (StemTherapy) at Lund University supported by Swedish Government and EU FP7 consortium TargetBrain. His research interests are generation and characterization of neural stem cell lines from different sources and development of stem cell-based treatments for stroke.

Jeffrey Kordower

Dr. Jeffrey H. Kordower is the Jean Schweppe-Armour Professor of Neurological Sciences, Professor of Neurosurgery, Director, Research Center for Brain Repair, Section Head of

Neuroscience, and Director of the Neuroscience Graduate Program at Rush University Medical Center. He received his B.A. and MA from the City University of New York and his Ph.D. in Neuropsychology from that same institution in 1984. He received an Honorary Doctorate of Science from that same institution in 2004.

Dr. Kordower is an international authority in the area of movement disorders, which special expertise in experimental therapeutic strategies in Parkinson's disease. He has published landmark papers in the area of cell replacement strategies including the first demonstration that fetal dopaminergic grafts can survive, innervate and form synapses in patients with Parkinson's disease, a paper that which was published in the New England Journal of Medicine. Furthermore, his recent demonstration that long-term grafts in such patients can form Lewy bodies was recently published in Nature Medicine. With regards to gene therapy, he published the lead article in science demonstrating that gene delivery of the trophic factor GDNF can prevent the emergence of motor symptoms and nigrostriatal degeneration in multiple nonhuman primate models of PD. A similar finding using gene delivery of neurturin has, in part, resulted in this therapy being tested in a Phase II clinical trials. He also was the first to demonstrate that gene delivery of trophic factors can obviate neurodegenerative processes in nonhuman primate models of Huntington's disease and Alzheimer's disease, with these studies being published in Nature and The Journal of Comparative Neurology, respectively.

Dr. Kordower has published over 350 papers, has lectured all over the world, and has been on over 20 Editorial boards. He is on the Scientific Advisory Boards of many biotech companies and scientific organizations. He is a Past-Councilor and Past President of the American Society for Neural Transplantation, Past-Chair for the Committee for the Use of Animals for the Society for Neuroscience, a member of the International Executive Committee for the Movement Disorders Society, and is a founding and current Scientific Advisory Board member, as well as former Executive Scientific Advisory Board Member for the Michael J. Fox Foundation.

Jia-Yi Li

Jia-Yi Li is Professor at the Wallenberg Neuroscience Center at Lund University in Sweden since 2011. He received a medical degree from Luzhou Medical College, China, in 1982; Master degree in West-China University of Medical Sciences (now Sichuan University) in 1988 and a Ph.D in neurobiology at University of Gothenburg, Sweden, in 1995. He led the first landmark study of Lewy pathology spread in PD patients who received neural transplantation. He is well acquainted with protein aggregation, propagation and neuronal dysfunction in the neurodegenerative diseases. His current research is focused on studying molecular and cellular mechanisms of propagation of Parkinson pathology and subsequent impaired neuronal function and induced cell death, aiming for novel therapeutics.

Meng Li

Dr. Li obtained her medical degree at Peking University and PhD from Edinburgh University. She established her research group in Edinburgh in 2000 with a prestigious MRC Career Development Award followed by a MRC Senior Non-Clinical Research Fellowship which she

spent at Imperial College London. She joined Cardiff University in 2012 as a professor of stem cell neurobiology.

Dr. Li's group is interested in elucidating the mechanisms underlining neuronal fate specification and applying these knowledge to devise novel strategies for driving pluripotent stem cell differentiation towards clinically important neurons, such as dopamine neurons, striatal GABAergic neurons and cortical interneurons. Using patient derived induced pluripotent stem cells (iPSCs) and genome edited hPSC models the group aims to uncover the cellular basis of neuropsychiatric diseases and to facilitate the development of cell therapies for neurological disorders.

Jeffrey Macklis

Jeffrey Macklis is the Max and Anne Wien Professor of Life Sciences in the Department of Stem Cell and Regenerative Biology, and Center for Brain Science, Harvard University, and Professor of Neurology [Neuroscience] at Harvard Medical School, and was founding Program Head, Neuroscience, Harvard Stem Cell Institute. He is an M.I.T. faculty member in the Harvard-Massachusetts Institute of Technology (M.I.T.) Division of Health Sciences and Technology. His lab is directed toward both: 1) understanding molecular controls and mechanisms over neuron sub-type specification, development, diversity, axon guidance-circuit formation, and degeneration in the cerebral cortex; and 2) applying developmental controls toward both brain and spinal cord regeneration and directed differentiation for in vitro therapeutic and mechanistic screening. The lab focuses on neocortical projection neuron development and sub-type specification; neural progenitor / "stem cell" biology; induction of adult cortical neurogenesis; subtype-specific axonal growth cone biology; and directed neuronal subtype differentiation via molecular manipulation of neural progenitors and pluripotent cells (ES/iPS). He is the recipient of a number of awards, including a Rita Allen Foundation Scholar Award, a Director's Innovation Award from the NIH Director's Office, The CNS Foundation Award, a Senator Jacob Javits Award in the Neurosciences from NINDS/NIH, The Cajal-Krieg Cortical Discoverer Prize, and he is an Allen Distinguished Investigator of the Paul G. Allen Family Foundation.

Malin Parmar

Malin Parmar is a professor at Lund University. Together with her lab she has shown in a series of high profile publications how human fibroblasts can be converted into neurons, how glial cells can be reprogrammed into neurons in vivo, and how functional dopamine neurons can be generated from human embryonic stem cells. She is a member of several European consortia, a founding member of the global network GForce-PD, and she has recently been awarded a prestigious grant from the European Research Council.

Rachael Pearson

Rachael Pearson graduated from Hertford College, University of Oxford in 1999 (Natural sciences: Physiological Sciences) before undertaking a PhD in retinal development with Professor Peter Mobbs in the Department of Physiology at University College London. During her PhD and first post-doctoral position, she became interested in the control of

retinal progenitor cell proliferation. This led fairly seamlessly into a post-doc with Professor Jane Sowden at the Institute of Child health, also at UCL, where Rachael began her work on stem cells and photoreceptor replacement therapy with Professor Sowden and Professor Robin Ali. In 2007, she was awarded a Royal Society University Research Fellowship and started her lab at the Institute of Ophthalmology. Since then, she has continued to work closely with Professors Ali and Sowden. Together, they have built a large programme of research developing stem cell based therapies for the treatment of retinal degeneration. Rachael was promoted to Reader (equivalent to Associate Professor) in Developmental Neuroscience in 2013. She is currently funded by the Royal Society, the MRC, Fight For Sight and the Alcon Research Institute.

Nils-Eric Sahlin

Nils-Eric Sahlin is a Professor (Chair) of Medical Ethics, Faculty of Medicine, University of Lund. He was formerly a Professor of Theoretical Philosophy at the same university. He is a fellow of The Royal Swedish Academy of Letters, History and Antiquities. Sahlin's main interests lie in decision theory and the philosophy of risk. He has developed theories of rational decision-making and epistemic risk-taking. He has also made contributions to the theory of evidence. Working closely with colleagues in Lund, he is currently finalizing papers on the following topics: the concept of epistemic risk, stem cells and risk, personalized, predictive and preventive medicine, and metaphysical explanations. Details of books Professor Sahlin has authored and edited can be found on his webpage: www.nilsericsahlin.se.

Jared Sternecker

Jared Sternecker is a group leader at the Center for Regenerative Therapies at the Dresden University of Technology. He received his PhD in 2005 from Johns Hopkins University in Baltimore, Maryland, in the laboratory of Prof. Dr. John Gearhart. Dr. Sternecker trained as a postdoctoral fellow in the department of Prof. Dr. Hans Schöler at the Max Planck Institute for Molecular Biomedicine, and from 2010 to 2014 was the project leader of the Stem Cell-Based Neurodegenerative Drug Discovery group. His group's previous and current research includes work on neurodegenerative diseases like amyotrophic lateral sclerosis (ALS) and Parkinson's disease (PD) and induced pluripotent stem cell (iPSC) technology.

Jun Takahashi

Jun Takahashi is Professor at Kyoto University Graduate School of Medicine. He studied medicine in Kyoto University, and took three years of residency in neurosurgery. He was given PhD from Kyoto University Graduate School of Medicine, and also passed the examination for Japanese Board of Neurological Surgeon in 1993. He started research on neural stem cells in Fred H. Gage's laboratory at the Salk Institute in La Jolla, where he worked as a postdoctoral fellow for two years. Since he came back to Kyoto in 1997, he has been working as a clinical neurosurgeon but continuing research work as well. Now he is a lecturer of the department of neurosurgery, performing mainly functional neurosurgery

including deep brain stimulation for Parkinson's disease patients, and also developing a method of cell transplantation using ES cells for the treatment of Parkinson's disease.

Håkan Widner

Håkan Widner is a Professor of neurology at Lund University and a clinical specialist in neurology with particular interest in Movement Disorders and complex and advanced neuroinflammatory disorders. He has been one of the original 5 who took neural tissue transplantation from the experimental phase to the clinic with particular responsibility of researching any immune responses against the grafted tissues. He did part of his PhD work in neurology in Lund and part at the Karolinska Institute at Clinical Immunology. He also investigated patients with toxin induced, MPTP induced parkinsonism and over several years worked with this project and experimental studies in non-human primates in California. Prof. Widner has been part of the NECTAR network from the beginning, served on its board, worked on the CAPIT/CAPSIT protocols and other committees over the years. He is currently involved in several clinical translational trials, foremost the TransEuro multicentre trial on embryonic neural tissue grafts.

PRACTICAL INFORMATION

Venue

AF BORGEN
Sandgatan 2
223 50 Lund
Tel: +46 (0)46 384 905



Speakers' accommodation

Grand Hotel
Bantorget 1
222 29 Lund
Tel: +46 (0) 46 280 61 00



Registration

The registration desk will be located on the bottom floor of AF Borgen, just in front of the conference room. Please follow the NECTAR signs to the conference room.

The registration desk will be open during the following hours:

Wednesday, December 9th	13:00 – 17:30
Thursday, December 10th	08:15 – 17:30
Friday, December 11th	08:00 – 15:30

At the registration desk you can collect your name badge and your program booklet. Please contact the conference staff throughout the day if you have any questions.

Please wear your name badge during the conference.

During the conference, the NECTAR meeting staff can be reached under the numbers:

+46 736 67 16 24 – Paulina Pettersson

+46 709 82 39 01 – Malin Parmar

Inside the plastic holder of your name badge you will find your tickets for the conference dinner(s) and the social activity taking place on December 9th, the latter for registered participants only. Please hand in those tickets to Paulina at the entrance to the event venue. If your plans change during the conference and you will NOT be able to attend the event you are registered to, you are welcome to return your event ticket to the registration desk or Paulina, who will then re-distribute them to guests placed on waiting lists to the event.

Conference dinner

The conference dinner on Thursday, December 10th is included in the delegate package. The dinner will be held at the Pelarsalen at the main University building, across from AF Borgen, at 19:00.

Should you have any dietary requirements and you have not specified them in the online registration form, please inform Paulina as soon as possible.



Lunch and coffee breaks

Coffee breaks & standing lunches will be served outside of the conference room.

Instructions for speakers

Considering the packed timetable of the meeting, we kindly ask speakers to present their talks within the allocated time.

PowerPoint projection facilities will be available, and technical assistance will be available throughout the meeting. We kindly ask all speakers to bring their talks on a USB-stick.

Oral Presentations (Datablitz sessions)

As in previous years, there will be Datablitz presentations: short 4-minute presentations followed by a 1-2 minute discussion. These will be timed by the Chair of the session and will not be allowed to overrun so we ask all speakers in these sessions to be particularly mindful of the time and the amount of data covered. We recommend that you do not try and present any more than 6 slides in your datablitz. One and the same laptop will be used for all datablitz presentations, which means that it will not be possible for you to present your datablitz from your own personal laptop. We therefore kindly ask all datablitz presenters to bring their talks on a USB-stick to the meeting and upload them on Paulina's computer well in advance of your presentation.

Considering the short timeframe of each presentation, the results will not be able to be discussed in detail, the presentations are however good springboards for discussions during the rest of the conference.

Official Language

The official language of the conference is English.

Wireless Network

AF Borgen offers complementary access to wireless internet in all the public areas and conference rooms. The available network is: AF Borgen Wifi, the password: SkP567!!

Accommodation

Delegates who have booked accommodation should deal with their hotels at the hotel desk.

Website: www.nectar-eu.net

Facebook: Find us on Facebook @ Nectar Pres

Twitter: Follow us on Twitter @NECTAR_EU, and Tweet using #NECTAR2015



TRAVEL WITHIN LUND

If you arrived to Lund by train, you arrived at Lund Central Station, situated just next to the city center. There are always a number of cabs available from a rank outside the station. Buses in Lund are either green or yellow. The green buses stay within Lund's city borders while yellow buses will leave Lund and go to e.g. Malmö. It is NOT possible to pay for your bus ticket onboard a bus with cash – you will need to use your credit card (yellow busses) or your mobile phone (both green & yellow busses) to pay for a single ticket. If you plan to take the bus a few times, you could also purchase a "Jojo Mini" card at Skånetrafiken's office at the Lund Central Station. Jojo Mini is a chip card you top-up with the amount of your choice - it will then allow you to travel easily with all of Skånetrafiken's buses (payment for a single ticket directly on the bus) and trains, including to and from Denmark (payment at a ticketing machine). You can top-up the Jojo Mini at Skånetrafiken's customer service centers as well as at ticketing machines.

The logo for Skånetrafiken, featuring the name in white text on a red rectangular background.

For schedules of trains and buses in Lund as well as in the whole region of southern Sweden and Copenhagen please visit www.sknetrafiken.se.

Rental bicycles – “LUNDAHOJ”



The most convenient way of moving around Lund is undoubtedly by bike. The company “Lundahoj” offers rental bikes which you pick-up and return to one of their many docking stations (see the picture). The first half hour of each ride is always free of charge, regardless of the number of trips per day. For a bike borrowed for longer than 30 minutes, the cost is 10 SEK for the first half hour, SEK 20 for second and after that 40 SEK per every

half hour. We recommend that you purchase a 3-day pass, which costs 25 SEK and can be bought directly at the bike stations with a credit card. A reservation sum of 140 SEK will be temporarily booked on your credit card for any charges exceeding the free 30 minutes. A good idea is to download the free app AllBikesNow! which will show you Lundahoj's all docking stations. For more information visit <http://lundahoj.se/>.

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Taxi Lund: www.taxilund121212.se

+46 (0)46 121 212

Taxikurir: www.taxikurir.se

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THE LEGEND OF ST. LUCIA

December 13th is the day when Swedes and others all over the world honor the legend of Saint Lucia. For many, many years Lucia has brought faith, hope, and a reason to believe in good things to come. Her legend stems from Syracuse on the island of Sicily. It is thought that during a time when the rulers of the land did not look favorably upon Christianity, a woman named Lucia had devoted her life to God and the poor. She gave her entire dowry to the poor, and thereby made the man she was to marry very upset. Lucia was put on trial, refused to renounce her Christian beliefs and was declared a witch. She was to be burned at the stake but when the guards tried to light the fire it would not light.

There are many theories on how the legend of Lucia came to Sweden. It could have been brought by priests, German traders or even by the Vikings in their adventures to southern

Europe. No one knows just how it evolved into the uniquely Swedish tradition it is today.



One popular version is a story of a terrible famine many years ago. On December 13th a well-lit ship on Lake Vänneren approached the shore carrying a woman at the helm dressed in white with a glow around her head. Having heard the Italian version the starving people thought it could be Saint Lucia coming to save them from this terrible famine.

The many Lucia songs all have the same theme:

*The night treads heavily / around yards and dwellings
In places unreached by sun / the shadows brood
Into our dark house she comes / bearing lighted candles,
Saint Lucia, Saint Lucia.* ♪ ♪ ♪ ♪ ♪ ♪

The Lucia celebrations include gingerbread and sweet, saffron-flavoured buns (see next page) shaped like curled-up cats with raisin eyes. You eat them with mulled wine or coffee.

Recipe for Lucia Buns (Lussekatte)



Ingredients

- 200 g unsalted butter, plus extra, to serve
- 450 ml milk
- 50 g fresh yeast, finely crumbled
- 1 egg, plus 1 extra, lightly beaten, to brush
- 165 g ($\frac{3}{4}$ cup) caster sugar
- $\frac{1}{4}$ tsp saffron threads, finely chopped
- 900 g (6 cups) plain flour
- 64 (about 85 g) raisins

Instructions

Preparation time: 1 hour and 25 minutes

Melt butter in a saucepan over medium heat. Add milk and heat until lukewarm. Pour mixture into a large bowl and add yeast, stirring to dissolve. Add egg, sugar, saffron and 1 tsp salt. Gradually add flour, stirring constantly, until mixture forms a smooth dough that comes away from the side of the bowl; don't worry if dough is sticky. Cover bowl with a clean tea towel and leave to rise in a warm, draught-free place for 45 minutes or until dough doubles in size.

Punch down dough and knead on a lightly floured work surface for 30 seconds or until smooth. Divide into 4, then divide each piece into 8. Shape each piece into a 20 cm length, then form into an S shape, tucking ends into dough to form an 8 shape, and pressing to join. Place on an oven tray lined with baking paper, cover with a tea towel and leave in a warm, draught-free place for another 40 minutes or until slightly risen.

Preheat oven to 200°C. Place a raisin into each circle created by the 8 shape, then brush with beaten egg. Bake buns for 10 minutes or until golden brown. Serve warm with butter.

SOCIAL ACTIVITY ON DECEMBER 9TH for registered participants only

Your ticket for the social activity can be found in your name badge holder - please bring it to the restaurant. If your plans change during the conference and you will NOT be able to attend the event, you are welcome to return your ticket to the registration desk or Paulina, who will then re-distribute them to guests placed on waiting lists to the event.

About the Restaurant Rauhrackel



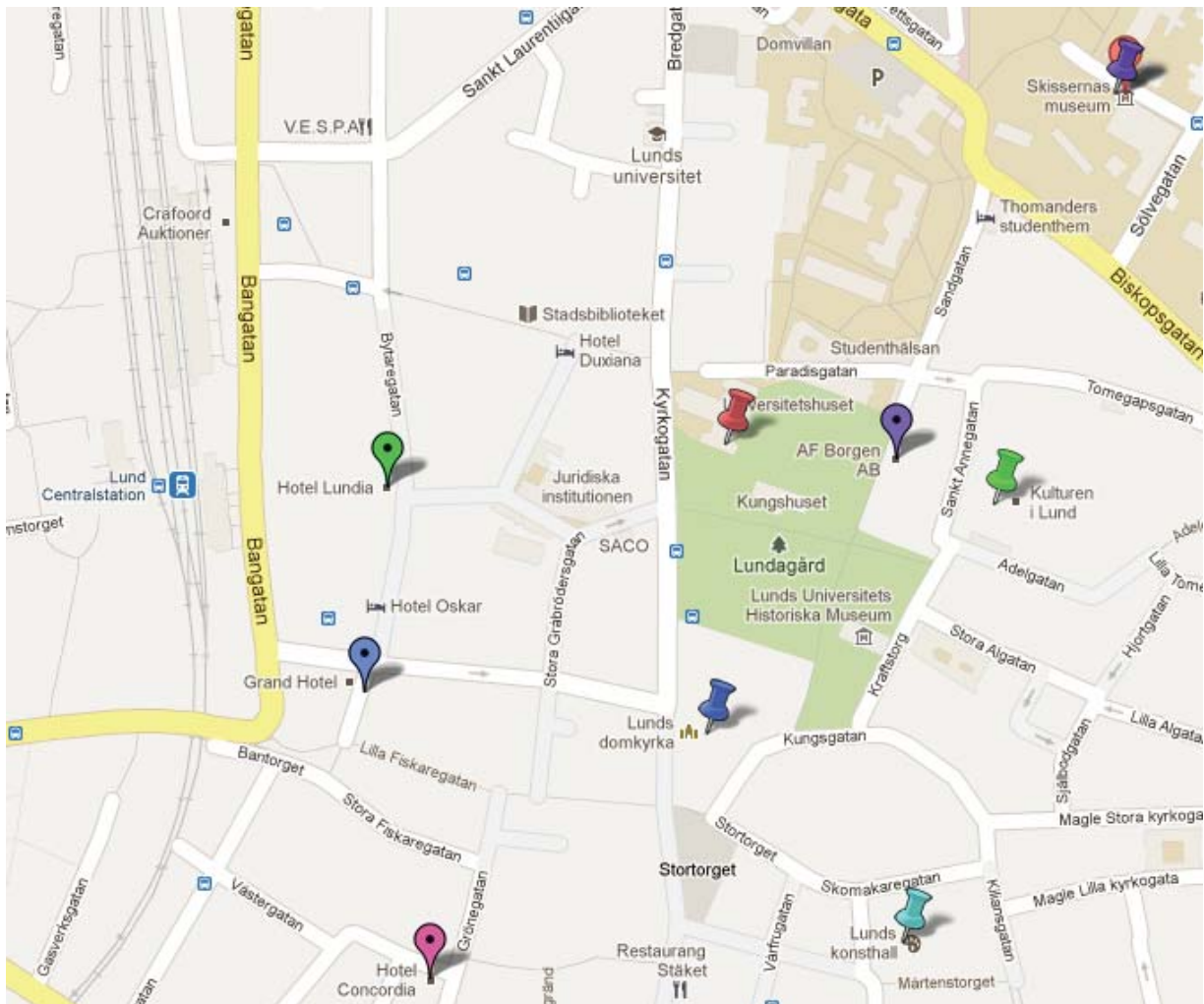
"Rauhrackel" is a fairytale creature, which according to the legend visited naughty children in Austria. In Lund "Rauhrackel" means something entirely different - it's (according to the restaurant owners) the coziest restaurant in the city. The restaurant's theme is the Tyrol, but inspiration is drawn from both the north and south of the region.










About the Beer Tasting

Your Beer Tasting begins with your host telling you about the origin of beer and about how beer has evolved to the modern product it is today. You will taste five kinds of beer of German & Austrian type (manufactured according to the German Purity Laws). These beers are: Lager, Pils, Weissbier, Märzen and Bockbier. Your host will tell you how the different kinds got created and explain the differences and similarities between them. You will also learn what to consider when choosing a specific beer kind to your meal. During the Beer Tasting you will also be served some snacks - cold cuts, cheese, pickles and Austrian pretzels.



MAP OF LUND



-  Grand Hotel
-  Hotel Lundia
-  Hotel Concordia
-  AF Borgen AB
-  Lund Cathedral
-  Lund University / Viking rune stones
-  Kulturen
-  Lund konsthall
-  The Museum of Sketches - Museum of Public Art

TOURIST ATTRACTIONS

In case you will be staying in Lund for a little longer or just need some fresh air to clear your thoughts, here are some tips of what to see.

Lund Cathedral



Lund Cathedral was built before 1085, the high altar of the crypt was consecrated in 1123. Its history gives a glimpse of the amazing history of Lund and it's the main tourist attraction in the city.

Address: Kyrkogatan, 222 22 Lund
Opening hours: 08:00-18:00 Mon-Fr,
09:30-17:00 Sat, 09:30-18:00 Sun,
no admission.

Lund University



The university is what makes Lund tick! It was founded in 1666, making it the

second-oldest university on Swedish ground. Today, it is one of northern Europe's most prestigious universities and one of Scandinavia's largest institutions for education and research. The university is spread around town, but some of the most beautiful buildings are located in Lundagård Park, not far from the cathedral and AF Borgen.

Viking rune stones



You'll find them in Lundagård Park surrounded by university buildings. They come from different villages in Skåne and tell stories of how a few Vikings met their death in different places in Northern Europe.

Kulturen



Kulturen is the second oldest open-air museum in the world. There are now more than thirty buildings filling two blocks of the city. The buildings show how

people have lived and worked in Sweden through the ages. You will also find some fifteen exhibitions about everything from peasants in Skåne to arts and crafts from all over the world.

Address: Tegérsplatsen, 223 50 Lund

Opening hours: 12:00 – 16:00 Tue – Sun.

Lund konsthall



Lund Konsthall is a contemporary art venue. They offer exhibitions and events

of high international standard, featuring art from the nearby region as well as from the rest of the world. They also engage in continuous research and experimentation with new exhibition formats.

Address: Mårtenstorget 3, 223 51 Lund,

Opening hours: mostly noon – 17:00, no admission.

The Museum of Sketches – Museum of Public Art



The Museum of Sketches was founded in 1934. Its focus is the creative process

around the conception of public art – that is to say sketches, preliminary works, and models to the art that meets us in our surroundings. It consists mainly of three main collections – the Swedish, the Nordic and the International collections.

Address: Finngatan 2, 223 62 Lund,

Opening hours: noon-17:00 Tue-Sun.

SPEAKERS DIRECTORY

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AWARED TRAVEL GRANTS **(alphabetical order)**

ANTONIO MARTIN-BASTIDA, Neurology Imaging Unit, Centre for Neuroinflammation and Neurodegeneration, Hammersmith Hospital Campus

CAROL NAUGHTON, Pharmacology & Therapeutics / Galway Neuroscience Centre, and School of Medicine, National University of Ireland, Galway

CHARLOTTE ERMINE, The Florey Institute of Neuroscience and Mental Health, University of Melbourne, Victoria, Australia

EMMA YHNELL, The Brain Repair Group Cardiff School of Biosciences, Cardiff University

ERIN DOLAN, Dept of Anatomy and Neuroscience, Western Gateway Building, Western Rd, UCC, Cork, Ireland

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ACKNOWLEDGEMENTS TO THE SPONSORS

We would like to acknowledge and thank the following agencies for sponsoring the event.

THE SWEDISH RESEARCH COUNCIL



DATABLITZ SESSIONS

Wednesday, December 9th, 2015

Datablitz - Session 1 16.45 – 17.40

Chair: Simon Stott

1	Andreas Heuer	Dopamine release from grafted primary rat VM transplants via optogenetic and chemogenetic stimulation
2	Carlos W. Gantner	Clinical-grade ventral midbrain dopamine neurons from human pluripotent stem cells for Parkinson's disease therapy
3	Patrick Aldrin-Kirk	Chemogenetic modulation of transplanted dopamine neurons reveals a novel serotonin-dependent Parkinsonian dyskinesia mechanism mediated by the 5-HT6 receptor
4	Niamh Moriarty	An injectable collagen hydrogel for the delivery of primary dopaminergic neurons into the Parkinsonian brain.
5	Stefanie Seilera	Neutralization of Nogo-A in the host brain improves graft function in a rat model of Parkinson's disease
6	Osama Elabi	Impact of Exendin-4 and Liraglutide on survival and efficacy of transplanted allogenic ventral mesencephalon cells in a rat model of Parkinson disease
7	Sara Nolbrant	Identifying predictive markers of successful graft outcome in animal models of Parkinson's disease through retrospective analysis of a large dataset of grafted cell preparations
8	Mariah Lelos	Can Stem-Cell Derived Dopamine Neurons Improve Non-Motor Impairments?

Thursday, December 10th, 2015

Datablitz - Session 2 10.00 – 10.50

Chair: Aideen Sullivan

1	Charlotte Ermine	Mechanisms regulating neurogenesis in the injured brain
2	Jonas Fritze	Dynamic Role of Chemokine receptor CXCR5 in Adult Neurogenesis
3	Janelle Drouin-Ouellet	Developing a protocol for efficient neuronal conversion of Parkinson's disease patient derived fibroblasts
4	Michaela Roth	Function and Mechanism of Pericyte Response after Experimental Stroke
5	Maria Pereira	Reprogramming cells in the brain - Assessing phenotype and tracing connectivity in vivo
6	Joanna Rzemieniec	3,3'-diindolylmethane protects neurons against hypoxia by targeting AhR-regulated CYP1A1
7	Philipp Berg	Alpha-Synuclein Proteostasis in Stem Cell-derived Cortical Neurons and its Role in Parkinson's Disease Dementia
8	Agnete Kirkeby	Modeling human neural tube patterning through culturing of stem cells under microfluidic gradients

Datablitz - Session 3 13.10 – 14.00

Chair: Mariah Lelos

1	Tiago Cardoso	Synaptic integration of transplanted human embryonic stem cell derived neurons in the adult rat brain
2	Daniel Tornero	Direct synaptic inputs from diverse host brain areas to grafted human iPSC-derived cortical neurons in stroke-injured rat cortex

3	Marie Jönsson	Comprehensive analysis of microRNA expression in regionalized human neural progenitor cells reveals microRNA-10 as a caudalizing factor
4	Síle M. Griffin	Regulation of Sox1GFP mouse embryonic stem cell neuronal differentiation by nicotinamide
5	Simon R W Stott	Temporal gene expression analysis of developing human midbrain dopamine neurons
6	Victoria Robertson	Characterising whole ganglionic eminence graft development
7	Weihua Li	Comparison of 11C-PE2I and 18F-DOPA PET for assessing progression and severity in patients with Early Parkinson's disease
8	Antonio Martin-Bastida	Striatal 11C-PE2I PET and Nigral Neuromelanin-sensitive MR in Parkinson's disease: a multimodal imaging study

Datablitz - Session 4 17.30 – 18.00

Chair: Rosemary Fricker

1	Fahad Somaa	Elevated but differential microglia responses on dopamine neurons survival and integration in different type of allografts in Parkinsonian mice
2	Ruth Concannon	Pronounced upregulation of the cannabinoid type-2 (CB2) receptor in neurotoxic, environmental and inflammation-driven rat models of Parkinson's disease
3	Cecilia Laterza	Role of glial scar in iPSC-derived It-NES behavior after transplantation in stroke-injured mice brain
4	Marco Barbariga	Immunomodulatory properties of pericytes in stroke
5	Francesca Stefani	Anti-tumoral effect of low-dose γ -irradiated mouse bone marrow-derived mesenchymal stromal cells

Friday, December 11th, 2015

Datablitz - Session 5 11.30 – 12.10

Chair: Daniella Rylander Ottosson

1	Erin Dolan	Characterisation of cognitive dysfunction in an α -synuclein model of Parkinson's disease
2	Carol Naughton	Intra-striatal administration of the environmental pesticide rotenone profoundly exacerbates motor dysfunction in the AAV- α -synuclein model of Parkinson's disease
3	Poonam Thakur	Increase in severity of α -synuclein pathology by addition of pre-formed fibrils
4	David J. Harrison	A behavioural characterisation of striatal quinolinic acid lesions in C57BL6/J mice as a model of neural transplantation in Huntington's disease
5	Kiah McCabe	Upregulation of Toll-like receptors in neurotoxic, environmental and inflammatory models of Parkinson's disease
6	Emma Yhnell	Cognitive training improves disease symptoms in a knock-in mouse model of Huntington's disease

Datablitz - Session 6 14.40 – 15.30

Chair: Marie Jönsson

1	Karolina Piracs	Overexpression of mutant huntingtin impairs macroautophagy
2	Heike Newland	Macroporous microcarriers for neural transplantation and growth factor delivery
3	Gang Wang	Massively Parallel in vivo Screen of Peptide Library Displayed on AAV2 for CNS-directed Gene Therapy

4	Alberto Martínez-Serrano	Electrophysiological properties of human neural stem cells grafted in an in vitro Parkinsonian model
5	Ludivine Breger	Unexpected immune response following the use of retrogradely transported lentiviral vectors in rats
6	Luis Quintino	Advanced inducible system to regulate neurotrophic factor gene therapy in PD
7	Marina Castro Zalis	Exploration of physical and chemical cues on retinal cell fate

DATABLITZ - SESSION 1: 16.45 – 17.30, Wednesday, December 9th, 2015

1.1 Andreas Heuer

Dopamine release from grafted primary rat VM transplants via optogenetic and chemogenetic stimulation

Authors: Andreas Heuer¹, Patrick Aldrin-Kirk², Bengt Mattsson³, Malin Parmar¹, Tomas Björklund², Martin Lundblad¹

Affiliations: 1: Developmental and Regenerative Neurobiology, Lund University, Lund, Sweden; 2: Molecular Neuromodulation, Lund University, Lund, Sweden; 3: Neurobiology, Lund University, Lund, Sweden

The generation of a “remote controlled stem cell transplant” would allow for fine-tuning of neurotransmitter release to potentiate the beneficial effects of the graft or to inhibit cell activity to avoid side effects. Recent developments in the fields of optogenetics and chemogenetics have made such manipulations possible via targeted expression of these receptors in distinct cell populations. In the present experiment we sought to express activating receptors in a rat ventral mesencephalic allograft paradigm. To achieve selective expression in the engrafted cells only we transplanted primary rat (TH::Cre knock in) VM cells into the denervated striatum of unilateral 6-OHDA lesioned rats. As only the transplanted cells possess Cre-recombinase we could selectively express our receptors of interest using flexed viral vectors. We chose to express either an activating light sensitive ion channel (Channelrhopsin 2) or an activating G-protein coupled chemogenetic receptor (hM3Dq), which can be activated via blue light or the otherwise inert ligand CNO, respectively. Subsequently we recorded the release of dopamine from the transplant using electrochemical detection methods using carbon fibre electrodes in vivo in anaesthetised rats directly at the site of the transplant. Our preliminary results reveal that both,

optogenetic and chemogenetic, approaches are viable to induce selective neurotransmitter release from the transplanted cells only.

1.2 Carlos W. Gantner

Clinical-grade ventral midbrain dopamine neurons from human pluripotent stem cells for Parkinson's disease therapy

Authors: Carlos W. Gantner^{1*}, Jonathan C. Niclis^{1*}, Walaa F. Alsanie¹, Stuart J. McDougall¹, Chris R. Bye¹, Andrew G. Elefanty^{2,3,4}, Edouard G. Stanley^{2,3,4}, John M. Haynes⁵, Colin W. Pouton⁵, Lachlan H. Thompson¹, Clare L. Parish¹

Affiliations: 1: The Florey Institute of Neuroscience and Mental Health, University of Melbourne, Melbourne, Australia; 2: Murdoch Children's Research Institute, The Royal Children's Hospital, Melbourne, Australia; 3: Department of Anatomy and Developmental Biology, Monash University, Clayton, Australia; 4: Department of Paediatrics, University of Melbourne, Melbourne, Australia; 5: Monash Institute of Pharmaceutical Sciences, Monash University, Melbourne, Australia.

* Equal contribution

Current pharmacotherapies provide only symptomatic benefit for Parkinson's disease patients. Cellular replacement therapy addresses the loss of ventral midbrain dopaminergic (vmDA) neurons, with clinical trials demonstrating proof-of-principle using fetal tissue. Human pluripotent stem cells (hPSCs) represent an alternative, sustainable tissue source for vmDA neuronal generation. However, existing differentiation protocols rely upon xenogeneic components that hinder clinical translation. Here we describe the first feeder- and xeno-free protocol for the generation of vmDA neurons from hPSCs. We utilise two novel reporter lines (LMX1A-eGFP and PITX3-eGFP) to demonstrate acquisition of OTX2/FOXA2/LMX1A precursors and FOXA2/TH/PITX3 neurons. Across multiple embryonic and induced hPSC lines our protocol increases the yield and proportion of mature vmDA neurons displaying classical functional properties. We identify the mechanism underlying these improvements and demonstrate high-throughput scalability and cryopreservation. Finally, transplantation of xeno-free vmDA progenitors into rodents demonstrates improved engraftment outcomes, confirming utility and providing critical steps towards clinical translation.

1.3 Patrick Aldrin-Kirk

Chemogenetic modulation of transplanted dopamine neurons reveals a novel serotonin-dependent Parkinsonian dyskinesia mechanism mediated by the 5-HT6 receptor

Authors: Patrick Aldrin-Kirk^{1,2}, Andreas Heuer^{1,2,3}, Gang Wang^{1,2}, Bengt Mattsson^{1,2,3}, Martin Lundblad^{2,3}, Malin Parmar^{2,3} and Tomas Björklund^{1,2}

Affiliations: 1: Molecular Neuromodulation, Department of Experimental Medical Science, Lund University, 221 84 Lund, Sweden; 2: Wallenberg Neuroscience Center, Lund University, 221 84 Lund, Sweden; 3: Developmental and Regenerative Neurobiology, Department of Experimental Medical Science, Lund University, 221 84 Lund, Sweden

Brain repair, through grafting of dopaminergic neuroblasts, has emerged over the past three decades as a possible alternative for the treatment of Parkinson's disease. Indeed, functional recovery was observed in patients following the pioneering clinical trials, however, therapeutic outcome has since been highly variable. In addition, a number of patients developed severe side effects, such as abnormal involuntary movements known as graft induced dyskinesias (GIDs), through an as of yet poorly understood mechanism involving serotonergic neurotransmission. In order to improve the therapeutic potential and investigate the mechanism underlying GIDs, we created a novel TH-Cre knock-in rat model, combining striatal dopaminergic cell replacement therapy with bimodal chemogenetic designer receptors exclusively activated by designer drugs (DREADDs). DREADDs are capable of modulating neuronal activity and transmitter release through G-protein coupled Gq, Gs and Gi pathways. Using this model, we demonstrated selective activation of only the dopaminergic neurons within the graft and that using bimodal chemogenetic DREADDs can significantly modulate and potentiate the therapeutic outcome. We further show that activating Gs-coupled DREADDs, leads to increased activity of adenylyate cyclase in dopaminergic neurons of the graft and is sufficient to induce GIDs in unprimed animals. Using this finding, we were able to establish a link between the activation of Gs-coupled 5-HT6 serotonergic receptors on the grafted dopaminergic neurons and the development of GIDs. These findings may be useful for designing future strategies for cell replacement therapy, increasing therapeutic outcomes and alleviating side effects in patients with Parkinson's disease.

1.4 Niamh Moriarty

An injectable collagen hydrogel for the delivery of primary dopaminergic neurons into the Parkinsonian brain

Authors: Niamh Moriarty¹, Abhay Pandit², E Dowd¹

Affiliations: 1: Pharmacology & Therapeutics and 2: CÚRAM Centre for Research in Medical Devices, National University of Ireland, Galway

The use of primary dopaminergic neurons derived from fetal ventral mesencephalon tissue as a routine therapeutic procedure is limited by poor graft survival with only ~5% of transplanted cells surviving the transplantation procedure. Biomaterial-based scaffolds have the potential to improve graft survival by providing a physical substrate into which pro-survival factors can be eventually complexed. Therefore, the aim of this study was to determine the neural cytocompatibility of a biomaterial scaffold composed of cross-linked collagen, and then to establish which level of crosslinking was most suitable to support cell survival in vivo. Injectable collagen hydrogels were fabricated from type 1 bovine collagen (2 mg/ml) and cross-linked with 4s-StarPEG (1, 2, 4, 6, 12 or 24 mg/ml). These were incubated with rat fetal ventral mesencephalon, primary astrocyte and mesenchymal stem cell (MSC) cultures for 48 hours, after which cell viability was assessed using alamarBlue® and LIVE/DEAD® assays. The effect on neurite outgrowth and dopaminergic viability was evaluated in ventral mesencephalon cultures using β III-tubulin and tyrosine hydroxylase immunocytochemistry respectively. To assess cell survival in vivo, GFP-MSCs encapsulated in hydrogels or vehicle (GFP-MSCs in transplantation media) were delivered bilaterally (30,000 cells in 3 μ l) into the striatum of male Sprague Dawley rats and were sacrificed on Days 1, 4 and 7 post surgery (n=4 per treatment, per timepoint). Graft survival was assessed using fluorescent microscopy, while collagen deposition and host immune response were assessed by collagen, GFAP and OX-42 immunohistochemical analysis. All collagen hydrogels were cytocompatible with each cell type and did not impede neurite outgrowth or negatively affect dopaminergic viability in ventral mesencephalon cultures. None of the hydrogels elicited an exaggerated host immune response, and successful GFP-MSC graft survival was found in hydrogels of lower 4s-StarPEG concentrations (1, 2 and 4 mg/ml). In conclusion, collagen hydrogels of all cross-linker concentrations were highly cytocompatible and hydrogels of lower 4s-StarPEG concentrations (1, 2 and 4 mg/ml) can be used for successful graft survival. This supports the hypothesis that collagen scaffolds could be used as a biomaterial scaffold to improve the outcome of reparative cell therapies in Parkinson's disease.

Acknowledgements: This work was funded through a Government of Ireland Postgraduate Scholarship from the Irish Research Council to Niamh Moriarty.

1.5 Stefanie Seiler

Neutralization of Nogo-A in the host brain improves graft function in a rat model of Parkinson's disease

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Transplantation of fetal ventral mesencephalic tissue into the striatum is a strategy to compensate for the characteristic dopamine deficits observed in Parkinson's disease. This therapeutic approach is, however, limited by the high numbers of fetuses needed associated with poor survival and suboptimal functional integration of the grafts in the host brain. Accumulating evidences indicate that contrasting inhibitory signals endowed in the central nervous system might support neuronal regeneration. Nogo-A is one of the most potent neurite outgrowth inhibitors in the central nervous system and its inhibition has been reported to result in functional recovery in several disease states. Hence, in the present study we aimed at improving survival and integration of grafted cells into the host brain by neutralizing Nogo-A. For that purpose, ventral mesencephalic tissue cultures were transplanted into the striatum of rats with unilateral 6-hydroxydopamine lesions. Concomitantly, rats received intraventricular infusions of neutralizing anti-Nogo-A antibodies for the first two weeks after transplanation. Control animals were similarly grafted and infused with control antibodies (IgG). Histological analyzes included number of graft-derived dopaminergic fibres growing into the host brain, numbers of dopaminergic neurons in the grafts as well as graft volumes. In addition, motor behavior by means of the cylinder test was assessed prior and after transplantation. Our results revealed that the group of rats infused with neutralizing Nogo-A antibodies showed a significant improvement of the asymmetrical forelimb use induced by the lesions. Importantly, we detected a significant three-fold higher number of graft-derived dopaminergic fibres growing into the host brain. Moreover, the antibody treatment resulted in a tendency for higher numbers of surviving dopaminergic neurons in the transplants. In sum, our findings support the view that inhibition of Nogo-A in the host brain may offer a novel and therapeutically meaningful intervention for cell transplantation approaches in Parkinson's disease.

1.6 Osama Elabi

Impact of Exendin-4 and Liraglutide on survival and efficacy of transplanted allogenic ventral mesencephalon cells in a rat model of Parkinson disease

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Introduction:

The glucagon-like peptide-1 receptor (GLP-1R) agonists exendin-4 and liraglutide have shown preclinical and clinical potential as neuroprotective therapeutics in the treatment of Parkinson's disease^{1,2,3}. Cell transplantation for the restoration of dopaminergic function has been hampered by low cell survival rates so we have hypothesised that the neuroprotective effects may support transplanted cells. Given that Parkinson's patients would typically be receiving L-DOPA prior to and following the cell transplantation procedure, the putative neuroprotective effects of these agents were tested both in the presence and absence of L-DOPA.

Methods:

Unilateral nigrostriatal dopaminergic neurons were lesioned by infusing 6-OHDA into the medial forebrain bundle of 7 groups of Sprague Dawley rats (n=8-9). Six groups received an intra-striatal infusion of cell suspension of ventral mesencephalic cells from e14 Wistar embryos. Three transplanted groups received chronic L- DOPA treatment prior to and following graft surgery. Paired groups of L-DOPA and non-L-DOPA groups received daily doses of exendin-4 (0.5 µg/kg twice daily) and liraglutide (100 µg/kg once daily) which started immediately after transplantation. Amphetamine rotation, stepping, whisker and cylinder tests were performed prior to and at regular intervals following cell transplantation. Brain, plasma, liver and adipose tissue samples were collected for further analysis 12 weeks post transplantation.

Results:

Exendin-4 significantly improved cell survival with superior behavioural amelioration in behavioural tests but these effects were lost in the presence of L-dopa. Liraglutide, however only improved cell survival and behavioural outcomes when administered with L-dopa. Plasma analysis illustrated developing hyperinsulinemia and hyperglycaemia in rats treated with exendin-4 and L-DOPA.

Conclusion:

GLP1R agonists have a supportive effect on cell survival and graft functionality. L-DOPA hindered the protective effect of exendin-4 possibly due to developing insulin resistance. Conversely L-DOPA potentiated the neuroprotective effect of liraglutide.

References:

1. Harkavyi A, et al., 2008 21:5-19
2. Li et al., 2015 Neurosci, 303: 42-50
3. Foltynie T and Aviles-Olmos 2014, 10(s1):38-46

1.7 Sara Nolbrant

Identifying predictive markers of successful graft outcome in animal models of Parkinson's disease through retrospective analysis of a large dataset of grafted cell preparations

Authors: Sara Nolbrant^{1,2}, Agnete Kirkeby^{1,2}, Katarina Tiklova^{3,4}, Shane Grealish^{1,2}, Andreas Heuer^{1,2}, Tiago Cardoso^{1,2}, Mariah Lelos⁵, Stephen Dunnett⁵, Thomas Perlmann^{3,4}, Malin Parmar^{1,2}.

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The prospect of using human embryonic stem cell (hESC)-derived mesencephalic dopaminergic (mesDA) progenitors as a cell source for neuronal replacement in Parkinson's disease is edging closer. However, a few final concerns need to be resolved before this therapy can be taken into clinical trials. Among these are the observed in vivo variations of the transplants in animal models, which hamper the possibility to accurately predict the grafting outcome. To address this issue we are performing a comprehensive retrospective analysis of grafting experiments performed using our cells at three different centers during the past five years, in order to re-evaluate the in vivo outcomes in terms of graft volumes, total tyrosine hydroxylase (TH)+ cell numbers, TH+ cell densities (i.e. TH+ cells/mm³) and functional characteristics. These observations are combined with RNA sequencing results from all the grafted cell preparations, with the aim of defining novel markers that can predict the grafting outcome. In this retrospective analysis, we have identified graft volume as the main contributor to the in vivo variation. Many of the genes that correlate with a large graft size were identified as belonging to a group of genes that are expressed at, or in close proximity to, the midbrain-hindbrain boundary. These genes play prominent roles in the development and maintenance of mesDA progenitors and neurons, thus indicating their contextual relevance. Importantly, some of the identified genes also correlate positively with the TH+ cell densities of the grafts, which suggest that they promote the acquisition of a dopaminergic phenotype. The identification of a panel of novel markers that can better predict a successful grafting outcome will be a valuable addition to the pre-clinical work of securing the safety and efficacy of the transplantable cell product and will bring hESC-derived cell replacement therapy one step closer to the patients.

1.8 Mariah Lelos

Can Stem-Cell Derived Dopamine Neurons Improve Non-Motor Impairments?

Authors: Lelos, M.J.¹, Fjodorova, M.², Kirkeby, A.³, Vinh, N.N.¹, Robertson, V.H.¹, Parmar, M.³, Li, M.², Rosser A.E.^{1,2}, Dunnett, S.B.¹

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Degeneration of the nigrostriatal pathway results in the manifestation of motor and non-motor symptoms in people with Parkinson's disease. Although it has been reported that the non-motor dysfunctions impact most heavily upon the quality of life for people with this disease, there are currently no interventions available to treat the non-motor symptoms. In a recent study, we found that human dopamine-rich fetal tissue (hVM) grafts are capable of alleviating both motor and non-motor dysfunctions, such as impaired motivation and visuospatial deficits. The aim of this project was to investigate whether more viable alternatives, stem cell-derived neurons, are also capable of improving non-motor dysfunctions. ES-derived dopamine neurons (ES-DA) were produced using two protocols and compared to human ventral mesencephalic tissue on a battery of simple motor tasks, as well as in an operant task measuring non-motor behaviours. The ES-DA neurons and hVM tissue alleviated simple motor functions over the 20 week post-graft period, but non-motor impairments were not improved. Although the ES-DA cells produced large grafts inundated with tyrosine hydroxylase positive neurons, and alleviated motor impairments, their maturation may be slightly delayed relative to hVM cells. We are only able to utilise the immunosuppressant cyclosporine A for up to 20 weeks, due to the adverse side effects. This, however, may be too short to observe changes in non-motor impairments. It is necessary to address the issue of immunosuppression to allow longer term, behavioural studies to reveal the capacity of novel stem cell-derived neurons to alleviate a larger range of more complex behavioural deficits.

DATABLITZ - SESSION 2: 10.00 – 10.50, Thursday, December 10th, 2015

2.1 Charlotte Ermine

Mechanisms regulating neurogenesis in the injured brain

Authors: Charlotte Ermine¹, Jordan Wright¹, Clare Parish¹, Lachlan Thompson¹

Affiliations: 1: The Florey Institute for Neuroscience and Mental Health, Australia

A key pathological feature of Parkinson's disease (PD) is the progressive degeneration of midbrain dopaminergic neurons, causing motor dysfunction. However there are a range of 'non-movement' related features (including cognitive dysfunction, dementia and sleep disorder), which are not alleviated by dopamine replacement therapy. We are currently investigating the hypothesis that reduced hippocampal neurogenesis contributes to cognitive dysfunction in PD. We aim to characterise the effect of the dopaminergic and noradrenergic system on the adult-hippocampal neurogenesis in order to identify potential targets for the treatment cognitive impairments related to neurogenesis.

We induced lesions of the different systems in adult rats using stereotaxic injections of toxins: 6-hydroxydopamine (dopaminergic system) and anti-dopa- β -hydroxylase-saporin (noradrenergic system). Four weeks later, the new cells were marked by pulses of bromodeoxyuridine (Brd-U) twice daily for 1 week. The animals were then sacrificed 4 weeks later for tissue collection.

A high-performance liquid chromatography has confirmed that both lesions were successful: dopamine level in the striatum dropped to 20% and noradrenaline level in the hippocampus dropped to 8.3%. Surprisingly there was no difference in the number of Brd-U positive cells or in the number of double positive Brd-U/NeuN cells between our groups.

The results show that while both noradrenergic and dopaminergic systems are implicated in the onsets of non-motor symptoms, they may not act through the regulation of adult-hippocampal neurogenesis like it was previously thought. Importantly our project has allowed reconsideration of how neurogenesis is involved in PD and redirected the therapies to better potential targets for treatment and brain restoration.

2.2 Jonas Fritze

Dynamic Role of Chemokine receptor CXCR5 in Adult Neurogenesis

Authors: Jonas Fritze, Nicole Huber and Henrik Ahlenius

Affiliations: Stem Cells, Aging and Neurodegeneration group, Lund Stem Cell Center, Department of Clinical Sciences, Lund University, Lund Sweden

The largest risk factor for developing neurodegeneration and cognitive decline is aging. Many neurodegenerative diseases appear together with impairment of neurogenesis and increased immune response. Chemokine receptor CXCR5 and its corresponding ligand CXCL13 are involved in organization and maintenance of B and T cells during inflammation. We recently showed an increased injury and density of pro-inflammatory M1 macrophages following stroke in CXCR5^{-/-} mice. Interestingly, CXCR5 have been suggested to regulate neurogenesis in adult zebrafish, and knockout of the receptor has been reported to increase the population of immature neural cells and decrease proliferation in the murine dentate gyrus (DG). However, how and if CXCR5 regulates neurogenesis in the aged brain is unknown.

We acquired in vivo data that, in contrast to previous studies, suggest an increased proliferation and decreased amount of neuroblasts in the subventricular zone (SVZ) of aged CXCR5^{-/-} mice. We also demonstrate expression of CXCR5 on SVZ derived NSPCs which is rapidly downregulated during differentiation, concomitantly with a dramatic increase of CXCL13 expression. Together our results propose that CXCR5 is involved in early NSPCs fate, and might be needed for proper neurogenesis. Better understanding of underlying mechanisms might give new insights in how neurogenesis is regulated by age related inflammation and its role in neurodegenerative disease.

2.3 Janelle Drouin-Ouellet

Developing a protocol for efficient neuronal conversion of Parkinson's disease patient derived fibroblasts

Authors: ¹Drouin-Ouellet J., ¹Pfisterer U., ¹Lau S., ²Collins LM., ²Barker RA. and ¹Parmar M.

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Direct reprogramming of somatic cells into specific neuronal subtypes has opened up the possibility not only to study disease processes in patient-specific neurons, but also to use these as a source of cells for brain repair therapies. We and others have successfully reprogrammed human fibroblasts into functional neurons, although conversion efficiency using adult human fibroblasts remains low. Here, using skin fibroblasts from 11 aged sporadic PD patients as well as 9 matched healthy subjects, we are comparing our current best protocol using previously identified factors (Ascl1, Brn2, Myt1L) as well as delayed transgene activation in combination with a dual SMAD signaling inhibition with new approaches such as the use of a new combination of small molecules, as well as methods to increase neuronal purity including magnetic (MACS) and fluorescence activated cell sorting. This study will help improve neuronal conversion efficiency of directly reprogrammed adult fibroblasts, which will be a step forward towards using these cells for cell-based replacement therapy as well as disease modeling of Parkinson's disease using patient-specific skin cells.

2.4 Michaela Roth

Function and Mechanism of Pericyte Response after Experimental Stroke

Authors: Michaela Roth⁽¹⁾, Ilknur Özen⁽¹⁾, Gesine Paul-Visse⁽¹⁾

Affiliations: (1) Department of Clinical Science, Lund University, Sweden

Pericytes, non-endothelial cells enclosed within the basement membrane of microvessels, have multifunctional activities, including contractile, immune, angiogenic functions and maintenance and control of the blood-brain-barrier. Disruption of these functions can lead to severe neurological defects and thus we are studying the role of brain pericyte under physiological and pathological conditions. For this purpose we use a mouse line that expresses GFP under the pericyte-specific promoter of Regulator of G-protein signaling 5 (Rgs5). Previously our lab has shown that pericyte are activated, leave the blood vessel wall and acquire a microglial phenotype after experimental stroke. However, the (1) function of these pericytes and (2) mechanism underlying these processes remain elusive.

First, we study the cellular function by characterizing the response of pericytes after experimental stroke in detail and address the question whether these cells are beneficial or detrimental for recovery after stroke. Preliminary data obtained by immunohistochemistry with M2 markers suggest that pericyte-derived microglial cells have an anti-inflammatory function.

Secondly, we investigate mechanisms of pericyte activation by studying the Rgs5 gene function. We are analyzing the response of heterozygous and Rgs5 knockout mice after stroke by comparing the vascular, immunological and cellular response in both mouse strains and thereby dissecting the gene function of Rgs5.

Our research will contribute to the understanding of pericytes and their potential use in a therapeutic approach.

2.5 Maria Pereira

Reprogramming cells in the brain - Assessing phenotype and tracing connectivity in vivo

Authors: Maria Pereira¹, Olof Torper¹, Malin Parmar¹

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Cellular reprogramming is a new and rapidly emerging field where somatic cells can be turned into pluripotent stem cells or other somatic cell types simply by expression of specific combinations of genes. By viral expression of neural fate determinants, it is possible to directly reprogram mouse and human fibroblasts into functional neurons, termed

induced neurons (iNs). Direct neural conversion can also be performed in vivo, where resident glia is reprogrammed into neurons.

Our group have previously shown that it is possible to convert resident striatal astrocytes into neurons in vivo. This was achieved via Cre expression under the control of a mouse GFAP promoter and using Cre-inducible lentiviral vectors expressing *Ascl1*, *Brn2* and *Myt1L* (ABM) (Torper et al., 2013). Since then, several groups succeeded at converting different types of cells in various brain regions, as well as in the spinal chord (Niu et al., 2013, Su et al., 2014, Heinrich et al., 2014). In a recent study, we show that adeno-associated viruses (AAVs) are efficient for neural conversion of both astrocytes and NG2-glia in vivo (Torper et al., 2015).

In this study, we explore different conversion factor combinations and how these affect the phenotype of the resulting induced neurons. We also trace connectivity of different cell types converted in the brain by making use of a modified rabbi virus that has recently been shown to work both for cells transplanted into the brain (Grealish, Heuer et al., 2015), in a host-to-graft or graft-to-host manner, and for cells reprogrammed in situ (Torper et al., 2015).

2.6 Joanna Rzemieniec

3,3'-diindolylmethane protects neurons against hypoxia by targeting AhR-regulated CYP1A1

Authors: Rzemieniec J., Litwa E., Wnuk A., Lasoń W., Kajta M.

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According to the latest statistics, stroke is the 5th leading cause of death worldwide. The lack of progress with respect to neuroprotective strategies against stroke has prompted scientists to search for novel and effective therapies. The most recent data indicated that protein expression and transcriptional activity of aryl hydrocarbon receptor (AhR) is increased in mouse cortical neurons after experimental stroke [Cuartero et al. 2014]. Nevertheless, there are no data concerning the protective capacity of selective aryl hydrocarbon receptor modulators (SAhRMs) e.g., 3,3'-diindolylmethane (DIM) in neuronal cells exposed to hypoxia. The aim of the present study was to investigate the neuroprotective potential of DIM against the hypoxia-induced damage in mouse hippocampal cells in primary cultures, with a particular focus on DIM interactions with the AhR and AhR-regulated CYP1A1. Our study demonstrated that DIM (0.1-10 μ M) in a concentration dependent manner inhibited hypoxia-induced loss of mitochondrial membrane potential, caspase-3 and lactate dehydrogenase activities. In AhR siRNA-transfected cells, DIM lost the anti-hypoxic capacity which pointed to AhR as a target of DIM-attributed neuroprotection. Indeed, DIM inhibited proteasomal degradation of AhR as evidenced by Western-Blot and nearly doubled AhR level at 6 h of hypoxia. This was

opposite to the effects of specific AhR agonists β -naphthoflavone and TCDD which induced proteasomal degradation of AhR and decreased its level starting at 3 h. Interestingly, long-term exposure to DIM reduced the AhR and CYP1A1 protein levels, which supports the antagonistic nature of this selective AhR modulator. Our data point to strong neuroprotective actions of DIM in hippocampal cells exposed to hypoxia. They also provide evidence that impairment of AhR and CYP1A1 plays a key role in the neuroprotective action of DIM against hypoxia-induced cell damage. This study may have implication for identifying new agents that could protect neurons against hypoxia by targeting AhR/CYP1A1 signaling.

The study was supported by statutory funds of the Institute of Pharmacology, Polish Academy of Sciences. Joanna Rzemieniec and Agnieszka Wnuk are holders of scholarship from the KNOW sponsored by Ministry of Science and Higher Education, Republic of Poland.

2.7 Philipp Berg

Alpha-Synuclein Proteostasis in Stem Cell-derived Cortical Neurons and its Role in Parkinson's Disease Dementia

Authors: Philipp Berg^{1,2,3}, Lewis DB Evans^{1,2}, Roger A Barker^{1,3} and Frederick J Livesey^{1,2}

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Approximately half of all Parkinson's Disease (PD) patients develop dementia within 10 years of their initial PD diagnosis, moreover in a familial form of early-onset PD, in which the alpha-synuclein gene (SNCA) is triplicated, all patients develop dementia. Histologically PD dementia is characterized by the presence of alpha-synuclein-containing Lewy bodies in neurons of the cerebral cortex and multiple lines of evidence suggest that the spreading of a pathological form of alpha-synuclein underlies disease progression. Little is known about the effect of increased alpha-synuclein levels on or how it might be transmitted between human cortical neurons.

Previously, we have combined methods for replaying human cerebral cortex development from pluripotent stem cells (Nature Neuro, 2012; Nature Protocols, 2012) with iPSCs derived from people with genetic forms of Alzheimer's disease to study early pathological changes in human excitatory glutamatergic neurons (Science Trans Med, 2012; Cell Rep, 2015).

We are using the same approach to model initiation and progression of PD dementia in cortical neurons, using iPSCs with a triplication of SNCA. Cortical neurons harbouring a SNCA triplication express three-fold higher levels of SNCA mRNA and alpha-synuclein protein compared to controls. Alpha-Synuclein is released from human cortical neurons in a cell-death independent manner and intracellular elevated alpha-synuclein levels in SNCA triplication neurons are accompanied by an increase in its release.

These cortical neurons provide a platform to study the cell autonomous effects of elevated alpha-synuclein, as well as the roles different cellular pathways play in alpha-synuclein release. This should help to understand how synucleinopathies establish and spread.

References:

- Moore S, et al., 2012: APP metabolism regulates tau proteostasis in human cerebral cortex neurons. *Cell Rep.* (5):689-96.
- Shi Y, Kirwan P, Livesey FJ. 2012: Directed differentiation of human pluripotent stem cells to cerebral cortex neurons and neural networks. *Nat Protoc.* (10):1836-46.
- Shi Y, et al., 2012: A human stem cell model of early Alzheimer's disease pathology in Down syndrome. *Sci Transl Med.* (124):124-29.
- Shi Y, Kirwan P, Smith J, Robinson HP, Livesey FJ. 2012: Human cerebral cortex development from pluripotent stem cells to functional excitatory synapses. *Nat Neurosci.* Feb 5;15(3):477-86.

2.8 Agnete Kirkeby

Modeling human neural tube patterning through culturing of stem cells under microfluidic gradients

Authors: Agnete Kirkeby¹, Marc Isaksson², Malin Parmar¹, Thomas Laurell²

Affiliations: 1: Wallenberg Neuroscience Center, Dept. Experimental Medical Sciences, Lund University; 2: Dept. Biomedical Engineering, Lund University.

Knowledge about structural brain development is almost purely derived from studies performed in rodents or smaller model organisms, despite the fact that the human brain is much more complex and 2000 times larger than that of the mouse. This research bias is caused by the impossibility of performing dynamic studies on anatomical brain patterning in human embryos, resulting in a significant lack of knowledge on human-specific neural development.

Here, we build a simplified 3D model of the developing human neural tube in vitro using human embryonic stem cells (hESCs). Taking advantage of established knowledge on neural tube patterning, we have designed a closed microfluidic culturing chamber for differentiation of hESCs. In this chamber, the cells are exposed to a gradient of chemicals during the first days of differentiation to mimic the anatomical gradients of growth factors present in the embryo around the developing neural tube. We show that hESCs survive well and differentiate into neural precursor cells when cultured in the gradient chamber. Importantly, through culturing in the gradient chamber we were able to induce progressive caudalization of neural identity, obtaining pure forebrain cells in the left side of the culture chamber to midbrain cells in the middle and hindbrain cells in the right side of the chamber, indicating an anatomical resemblance to the rostro-caudal organization of the neural tube.

Remarkably, we found the gene WNT1 to be very highly expressed in the area of the midbrain-hindbrain boundary in the culture chamber, indicating the formation of an in vitro equivalent to the anatomical structure of the Isthmic Organizer (IsO).

Our model will be a unique and novel tool for studying important signaling between neighboring neural cell populations during development, and for investigating how human brain development differs from that of other species in order to achieve an extraordinary degree of complexity.

DATABLITZ - SESSION 3: 13.10 – 14.00, Thursday, December 10th, 2015

3.1 Tiago Cardoso

Synaptic integration of transplanted human embryonic stem cell derived neurons in the adult rat brain

Authors: Tiago Cardoso^{1,2}, Andreas Heuer^{1,2}, Agnete Kirkeby^{1,2}, Shane Grealish^{1,2}, and Malin Parmar^{1,2}

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Human embryonic stem cell (hESC)-derived neurons patterned towards a midbrain dopaminergic fate have been shown to survive long-term, release dopamine and to extensively innervate correct host structures after transplantation into the central nervous system. Using a rabies-based monosynaptic tracing technique, we have recently shown that hESC-derived neurons are able to integrate into host circuitry as early as 6 weeks post-transplantation by establishing both host-to-graft and graft-to-host synaptic connectivity. We are now using the same tracing methodology to investigate the connectivity of cells transplanted to the striatum and to the substantia nigra. In this work, 6-OHDA lesioned rats received intrastriatal or intranigral transplants of hESC-derived neurons. Then, to assess for synaptic connectivity, animals were injected with rabies vector either 5 or 17 weeks after transplantation and perfused one week later for histological analysis.

Using this strategy we show that host neurons in local structures as well as distant afferent structures such as prefrontal cortex, striatum, motor cortex, thalamus, substantia nigra and raphe nucleus are able to establish synaptic connections with neurons transplanted in either forebrain or midbrain. This pattern was observable at an early time point of 6 weeks post grafting and maintained at 18 weeks. Moreover, when hESCs patterned towards a forebrain or midbrain fate were grafted in a heterotopic manner (i.e., into substantia nigra or striatum respectively), the capacity of these neurons to integrate host circuitry was retained. Overall, we show that both homotopic and heterotopic hESC-derived grafts can integrate into host circuitry. A better comprehension on graft-host neuronal communication might be fundamental towards bringing cell replacement therapy towards successful clinical application.

3.2 Daniel Tornero

Direct synaptic inputs from diverse host brain areas to grafted human iPSC-derived cortical neurons in stroke-injured rat cortex

Authors: Daniel Tornero¹, Oleg Tsupykov², Cristina Rodriguez¹, Somsak Watanabe¹, Jemal Tatarishvili¹, Shane Grealish³, Malin Parmar³, Galina Skibo², Olle Lindvall¹ and Zaal Kokaia¹.

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Brain repair after damage represent a major challenge for current clinical and basic research. Recently, we have shown that transplantation of human skin-derived neuronal precursors is a good strategy to improve functional recovery following cortical stroke in a rat model. Grafted cells give rise to mature neurons that re-build the damaged tissue and send fibers to several host structures. However, there are still unanswered questions regarding the mechanism underlying the functional integration of grafted cells into the neuronal network of the injured brain. Regeneration of functional synaptic connections between host neurons and transplanted cells is of crucial importance in order to accomplish cell replacement, which would lead to long-term recovery after stroke.

Our last results demonstrate that human induced pluripotent stem (iPS)-derived cells grafted into the stroke-damaged rat brain receive synaptic inputs from host cells in a very specific manner, sometimes from structures located several millimetres away from the implantation site. Immuno-electron microscopy (iEM) revealed synaptic contacts between host neurons and the neurons generated from grafted cells; and monosynaptic tracing of afferent connections with modified rabies virus showed a pattern that resembles that of the local endogenous neurons lost during stroke damage. These data clearly show that human iPS-derived cells transplanted into the stroke damaged cortex, beyond cell survival and differentiation, can integrate into the host network. Our results provide insight into the therapeutic potential of stem cell transplantation for stroke damage in the adult brain.

3.3 Marie Jönsson

Comprehensive analysis of microRNA expression in regionalized human neural progenitor cells reveals microRNA-10 as a caudalizing factor

Authors: Jönsson E Marie¹, Nelander Wahlestedt Jenny², Åkerblom Malin¹, Kirkeby Agnete², Malmevik Josephine¹, Brattås L Per¹, Jakobsson Johan¹, Parmar Malin²

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MicroRNAs (miRNAs) have been implicated in regulating multiple processes during brain development in various species. However, the function of miRNAs in human brain development remains largely unexplored. Here, we provide a comprehensive analysis of miRNA expression of regionalized neural progenitor cells derived from human embryonic stem cells and human foetal brain. We found miR-92b-3p and miR-130b-5p to be specifically associated with neural progenitors and several miRNAs that display both age-specific and region-specific expression patterns. Among these miRNAs, we identified miR-10 to be specifically expressed in the human hindbrain and spinal cord, while being absent from rostral regions. We found that miR-10 regulates a large number of genes enriched for functions including transcription, actin cytoskeleton and ephrin receptor signalling. When overexpressed, miR-10 influences caudalization of human neural progenitor cells. Together, these data confirm a role for miRNAs in establishing different human neural progenitor populations. This dataset also provides a comprehensive resource for future studies investigating the functional role of different miRNAs in human brain development.

3.4 Síle M. Griffin

Regulation of Sox1GFP mouse embryonic stem cell neuronal differentiation by nicotinamide

Authors: Síle M. Griffin¹, Mark R. Pickard¹ and Rosemary A. Fricker¹

Affiliations: 1: Institute for Science and Technology in Medicine, Keele University, Staffordshire, UK.

Until recently very little research has been undertaken into the role of nicotinamide (vitamin B3 metabolite) during early neural development. We previously reported the important functioning of nicotinamide as a regulator of neuronal differentiation and maturation in mouse embryonic stem cell (mESC; 46C Sox1GFP reporter cell line) monolayer cultures.

Further, mESCs differentiated in serum-free medium with nicotinamide (10 mM) alone showed enhanced conversion to catecholaminergic neurons at the expense of other neuron subtypes.

The first aim of this study was to examine the mechanisms by which cells change their fate, by evaluating the effects of nicotinamide on cell division / cell cycle exit and pro / anti-apoptotic mechanisms. Secondly, the potential of nicotinamide was combined with external signalling factors known to enhance a dopaminergic phenotype and immunocytochemistry/fluorescence microscopy was performed to assess the extent of differentiation.

Novel findings highlighted a reduction in the proportion of proliferating cells in nicotinamide-treated cultures – nicotinamide suppressed Sox1 precursor proliferation, thereby driving the proliferation to differentiation switch during development and promoting neuronal differentiation. Crucially, treatment with nicotinamide had no adverse effects on cell viability of monolayer mESC cultures with no increase in cell death observed. Findings outlined in this study are exciting, as nicotinamide uniquely appears to drive dopamine neuron differentiation as effectively as known cocktails of signalling factors routinely applied in current published protocols.

The strong clinical potential of nicotinamide means that it could successfully be applied to future neural cell-based therapies as a valuable component of cell culture media to aid the conversion of stem cells to dopamine neurons; helping to increase the efficiency of this process, the safety of the cells produced, and reduce costs; as we progress towards a patient-specific cell replacement therapy.

The authors acknowledge Professor Adrian Williams, QEHB and Professor Clive Hawkins, UNHS for financial support.

3.5 Simon R W Stott

Temporal gene expression analysis of developing human midbrain dopamine neurons

Authors: Simon R W Stott¹, Enrique Toledo², Emmanouil Metzakopian³, Ernest Arenas², & Roger A Barker¹

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A more complete understanding of the transcriptional programme involved in midbrain dopamine neuron development would be of great value in efforts to generate these cells in vitro for the purpose of cell transplantation. To this end, we have conducted RNA sequencing analysis of tissues dissected from three regions of the developing human brain: the diencephalon, mesencephalon and metencephalon. Dorsal, lateral and ventral sub-

dissections were made from each of these three regions, with the ventral mesencephalon being further divided into rostral and caudal portions. RNA extracted from these sub-dissections was sequenced allowing for comparative analysis to be conducted. Two different time points (8 weeks and 10 weeks post conception) have been analysed in this manner. In situ hybridization of differentially expressed genes has highlighted interesting new expression patterns. When comparing the ventral midbrain floorplate area with the rest of the dissected regions, many well recognised dopamine-related genes were apparent (such as ALDH1A1, TH, DDC, EN1, PITX3, EN2, NTN1, FOXA2, FOXA1, & SHH). Many interesting novel genes, however, were also encountered, such as SCUBE1 (signal peptide, CUB domain, EGF (epidermal growth factor)-like protein) which encodes a glycoprotein involved in sonic hedgehog functioning. The expression pattern of Scube1 is limited to the ventricular layer of the floorplate region in the developing midbrain. Another gene of interest is PEG10 (Paternally Expressed Gene 10), which is a Retrotransposon-derived protein that is known to inhibit TGF-beta signaling and prevent apoptosis via interactions with SIAH1. In the developing human midbrain, Peg10 is expressed by the dopamine neurons. This work conducted in human tissue is now being compared with gene expression patterns in the developing mouse midbrain to determine differences.

3.6 Victoria Robertson

Characterising whole ganglionic eminence graft development

Authors: Robertson VH, Harrison C, Lelos MJ, Potter H, Vinh NN, Torres EM, Rosser AE, Dunnett SB

Affiliations: Brain Repair Group, School of Biosciences, Cardiff University, Cardiff, Wales

Transplantation of cells derived from human primary foetal whole ganglionic eminence (hWGE) can improve function in Huntington's disease (HD). Due to practical limitations of using hWGE for therapeutic purposes, there is much interest in generating DARPP-32 positive medium spiny neurons (MSNs) from pluripotent stem cells. Before clinical use, cells must be shown to be safe and functional following transplantation into animal models of HD. However, despite functional improvements following transplantation of rodent WGE into rodent models of HD, and preliminary evidence that hWGE grafts improve function in HD patients, few reports have shown convincing functional recovery following transplantation of hWGE or human stem cell-derived MSNs into rodent HD models. There are several possible reasons for this; including the slower development of human foetal cells, demanding extended periods post-transplantation to detect functional recovery. The lack of a clear functional benefit from hWGE transplants is impeding efficient preclinical assessment of novel human donor cells. We therefore aimed to systematically characterise the development of transplanted hWGE cells in a rat model of HD in comparison to rat WGE transplants, which are currently better understood.

Unilateral grafts were transplanted into the quinolinic acid lesioned rat striatum. To avoid immune rejection of transplanted human xenografts, we used either daily cyclosporine injections or neonatal desensitisation. We report histological assessment of rat and human WGE transplants at 2, 4, 8 and 16 weeks post-transplantation. Early time-points show the presence of proliferating cells, developing into a mature neuronal phenotype over time. Grafts are seen to integrate into the host brain over time and are innervated with host dopaminergic fibres positive for tyrosine hydroxylase, coinciding with an increase in DARPP-32 positive medium spiny neurons within the graft. These data will provide a benchmark upon which to compare the development of grafts derived from pluripotent stem cells.

Acknowledgements

Funding through “Repair-HD”, an EU FP7 grant.

3.7 Weihua Li

Comparison of 11C-PE2I and 18F-DOPA PET for assessing progression and severity in patients with Early Parkinson’s disease

Authors: Li W. ¹, Lao-Kaim N. ¹, Roussakis A. ¹, Martin-Bastida A. ¹, Politis M. ^{1,2}, Valle-Guzman N. ³, Kefalopolou Z. ⁴, Paul G. ⁵, Widner H. ⁵, Foltynie T. ⁴, Barker R. ³, Piccini P. ¹

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Background:

Positron emission tomography (PET) with 18F-DOPA has long been regarded as the ‘gold standard’ for assessing the progression in vivo in Parkinson’s disease and for monitoring the efficacy of neuroprotective treatments in clinical trials. 18F-DOPA uptake reflects the activity of aromatic amino acid decarboxylase (AADC), and the storage capacity of 18-Fluorodopamine. However, previous studies have reported that 18F-DOPA PET overestimates striatal dopaminergic nerve terminal density in early Parkinson’s disease (PD) patients. 11C-PE2I is one of the most recent PET radioligands with high affinity and selectivity for the dopamine transporter (DAT). We hypothesise that 11C-PE2I may be a good alternative to 18F-DOPA for assessing disease progression and changes related to neuroprotective treatments in patients with PD.

Objective:

To compare 18F-DOPA with 11C-PE2I PET, and then demonstrate their ability in assessing the severity and rate of progression of PD.

Materials and Methods:

The striatal uptakes of 18F-DOPA and 11C-PE2I were compared using PET in 31 PD patients (age mean±SD: 55.46 ± 7.09 years; disease duration mean±SD: 5.7 ± 2.2 years; UPDRS

mean±SD: 32.06 ± 11.63) recruited into the Transeuro study. The correlation of uptakes with UPDRS-III total and sub-scores was also investigated.

Results:

As expected there was a significant negative correlations between 18F-DOPA uptake and total UPDRS-III score in bilateral caudate ($r = -0.37$, $p = 0.04$) and putamen ($r = -0.40$, $p = 0.02$), However, the negative correlation between 11C-PE2I uptakes and total UPDRS-III in bilateral caudate and putamen was even more robust ($r = 0.51$, $p = 0.003$ and $r = -0.51$, $p = 0.004$ respectively)

Conclusions:

11C-PE2I PET measures more accurately the loss of dopaminergic nerve terminals than 18F-DOPA and could be appropriate to assess neuroprotective treatments in PD.

3.8 Antonio Martin-Bastida

Striatal 11C-PE2I PET and Nigral Neuromelanin-sensitive MR in Parkinson's disease: a multimodal imaging study

Authors: Martin-Bastida A.¹, Lao-Kaim N.¹, Roussakis A.¹, Politis M.^{1,2}, Li W.¹, Valle-Guzman N.³, Kefalopoulou Z.⁴, Paul G.⁵, Widner H.⁵, Foltynie T.⁴, Barker R.³, Piccini P.¹.

Affiliations: 1: Neurology Imaging Unit, Imperial College London, UK; 2: Neurodegeneration Imaging Group, Kings' College London, UK; 3: Brain Repair Centre, Cambridge University, UK; 4: Unit of Functional Neurosurgery, University College London, UK; 5: Translational Neurology Group, Lund University, Sweden.

Background

The main pathological features in Parkinson's disease (PD) result from the progressive loss of pigmented dopaminergic neurons in substantia nigra compacta (SNc) and the subsequent reduction of dopaminergic striatal afferences. Positron Emission Tomography (PET) with 11C-PE2I has high affinity for striatal dopamine transporter (DAT) when compared with other traditional PET tracers as 18F-DOPA. Furthermore, analysis of Neuromelanin (NM) contrast of SNc with high resolution T1 weighted (T1w) MR allows distinguishing PD and healthy controls with high accuracy. Here we combined striatal BPND quantification with PE2I PET and Nigral NM-contrast T1 in the same individuals to explore PD patients' nigrostriatal pathway.

Methods

Sixteen patients with PD recruited into Transeuro study were scanned with 11C-PE2I PET, NM T1 weighted and 3D MPRAGE anatomical MR scans along with 12 healthy volunteers age-matched. Pre-processing and kinetic modelling for the 11C-PE2I PET data was conducted using MIAKAT. Brain extraction was performed on the MPRAGE images segmented and registered to Montreal Neurological Institute (MNI) template using the Anterior Commissure as anatomical reference. Bilateral delineation of ROIs 10 mm² for the

SNc and SCP were respectively performed and contrast ratios (CRs) calculated of the medial, central, and lateral parts of the SNc. Contrast ratios weighted MR were compared between PD and healthy controls using non-parametric Wilcoxon's rank test. Striatal PET and nigral MR data were correlated using Pearson coefficient.

Results

PD patients presented reduced CR of lateral SNc ($p < 0.05$) when compared with healthy controls. Significant correlations were found between SNc central and Pre-commissural striatum, both ventral and dorsal caudate and putaminal nuclei. SNc medial correlates weakly with pre-commissural ventral caudate. No correlations were found with SNc lateral.

Conclusion

The absence of significant correlations between posterior striatum and SNc lateral supports the nigrostriatal lateral-dorsal gradient of neurodegeneration in PD.

DATA BLITZ - SESSION 4: 17.30 – 18.00, Thursday, December 10th, 2015

4.1 Fahad Somaa

Elevated but differential microglia responses on dopamine neurons survival and integration in different type of allografts in Parkinsonian mice

Authors: F. Somaa, J.A. Kauhausen, L.H. Thompson, C.L. Parish

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Post-mortem evaluation of human fetal tissue grafts in Parkinsonian patients have shown the presence of numerous immune cells including microglia, macrophages, T and B cells. However it remains unclear whether the sum total of these immune cells plays a net protective or detrimental role. We therefore sought to examine the inflammatory response following VM implants on graft survival and integration, and to understand the role of these innate immune cells specifically in the absence of adaptive immune response. Mice received 6-hydroxydopamine midbrain lesions 1 week prior to intrastriatal allografts or isografts of ventral midbrain fetal tissue. To assess the impact of the inflammatory response on grafts, TH-GFP C57BL/6 tissue was implanted into C57BL/6 mice (BL6-BL6), Swiss mice (BL6-SW), and into nude mice (BL6-NUDE). In addition tissue from TH-GFP Swiss mice was grafted into Swiss (SW-SW) and C57BL/6 mice (SW-BL6). At 6 weeks post-transplantation, quantification of GFP+ cells revealed the largest grafts in nude animals followed by isogenic grafts, with allografted animals showing the poorest survival. Similarly graft integration was enhanced in isografts and greatest in nude recipients, compared to allografted animals. These findings were supported by an increase in reactive astrocytes in allografted animals. Surprisingly, reactive microglia were elevated in both allografted and nude recipients. Further examination revealed differential microglial responses with allografted animals showing an increase in pro-inflammatory M1 microglia (CD16/32) accompanied by T-cell infiltration

(CD3), while microglia surrounding grafts in nude mice were predominantly of M2 (CD206) neurotrophic phenotype, and devoid of T-cells. In summary, this study suggests new mechanisms of allograft rejection in PD, specifically through an effect of T-cell on microglia activity. Our findings suggest that T cells interact with microglia and result in immune signaling that is strongly neurotoxic (M1), yet in the absence of T-cell infiltration microglia display a neurotrophic phenotype (M2). Our finding suggest greater attention to T cell responses, by way of immune-suppressing drugs, are required to modulate microglia responses and promote optimal transplantation outcomes.

4.2 Ruth Concannon

Pronounced upregulation of the cannabinoid type-2 (CB2) receptor in neurotoxic, environmental and inflammation-driven rat models of Parkinson's disease

Authors: R.M. Concannon^{1,2}, B.N. Okine^{1,2}, D.P. Finn^{1,2}, E. Dowd^{1,2}

Affiliations: 1: Pharmacology & Therapeutics and 2Galway Neuroscience Centre, National University of Ireland, Galway

Neuroinflammation may contribute to the progression of neurodegenerative disease, forming a 'self-sustaining cycle' of neurodegeneration and neuroinflammation in conditions such as Parkinson's disease (PD). As such, anti-inflammatory therapy for restoration in neurodegenerative disease has emerged, and one potential target is the immune-suppressive microglial cannabinoid type-2 (CB2) receptor. However, in order to assess the preclinical neuroprotective efficacy of drugs targeting this receptor, its expression in animal models of PD needs to be determined. Therefore, the aim of this study was to investigate and compare the changes that occur in the endocannabinoid system in neurotoxic, environmental and inflammation-driven models of PD.

Male Sprague Dawley rats were given a single intra-striatal injection of 6-OHDA (10 mg), rotenone (1.25 mg), LPS (10 mg) or Poly I:C (20 mg), with corresponding vehicle on the other side. Animals underwent behavioural testing for motor dysfunction on Days 7, 14 and 28 days, and were sacrificed on Days 1, 4, 14 and 28 post-surgery (n=8 per treatment, per timepoint). Changes in the endocannabinoid system were investigated by qRT-PCR, mass spectrometry and immunohistochemistry.

Following injection of direct dopaminergic and indirect inflammatory toxins into the rat striatum, the CB2 receptor was significantly elevated across all models which correlated significantly with an increase in microglial activation. Interestingly, the increase in CB2 receptor expression in the inflammation-driven models was significantly more pronounced than that in the neurotoxic models. In addition, elevation of striatal endocannabinoid levels was also observed following LPS injection, a change that was not observed in any of the other models.

Thus, this study has shown that the endocannabinoid system is dysregulated in different models of Parkinson's disease, and has also revealed significant differences between the

models themselves. This study indicates that targeting the CB2 receptor may represent a viable target for anti-inflammatory disease modification in Parkinson's disease.

Acknowledgements: The authors gratefully acknowledge the support of Health Research Board Grant no. (HRA_POR/2012/12).

4.3 Cecilia Laterza

Role of glial scar in iPSC-derived It-NES behavior after transplantation in stroke-injured mice brain

Authors: Laterza C.¹, Hara N.¹, Ge R.¹, Tornero D.¹, Pekny M.², Lindvall O.¹ and Kokaia Z.¹

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Astrocytes are a key cell type playing active role in physiology and pathology of the CNS. They use an intermediate filament network as a signalling platform and a structural scaffold to coordinate the appropriate responses. After stroke, besides cell death in the ischemic core, there are many alterations in the penumbra area, among which reactive astrogliosis and glial scar formation play essential roles. The reactivity of astrocytes after stroke has both detrimental and beneficial effects. The capacity of reactive astrogliosis in limiting the tissue damage at the acute phase is balanced against restricted regenerative potential later on.

In this project we aim to evaluate the influence of glial scar on the therapeutic potential of human iPSC-derived neuroepithelial-like stem (It-NES) cells after their transplantation in stroke-injured mice brain.

To pursue our aim we used a transgenic mouse lacking both Vimentin and GFAP. Vim^{-/-}/GFAP^{-/-} mice show impaired astrocyte activation and glial scar formation upon CNS injury. We induced cortical lesion by occlusion of distal branch of middle cerebral artery both in Vim^{-/-}/GFAP^{-/-} and WT mice and transplanted 1x10⁶ It-NES cells close to the penumbra. Two months after transplantation the mice were sacrificed and brains were analyzed. Our preliminary data confirmed the absence of glial scar formation in Vim^{-/-}/GFAP^{-/-} mice. We detected some GFAP-positive cell in Vim^{-/-}/GFAP^{-/-} mice, but they were exclusively coming from grafted cells. Moreover, we showed that the absence of glial scar did not affect the lesion size. We are currently analyzing the survival, proliferation and integration of transplanted It-NES cells in Vim^{-/-}/GFAP^{-/-} mice to assess how glial scar is affecting the properties of grafted cells.

Our data will shed light on the interplay between astrocytes and It-NES cells, contributing to the improvement of cell-based treatment strategies, both in promoting brain repair and in reducing neurological impairment after stroke.

4.4 Marco Barbariga

Immunomodulatory properties of pericytes in stroke

Authors: Marco Barbariga, Ilknur Özen, Abderahim Gaceb, Gesine Paul-Visse

Affiliations: Translational Neurology Group, Department of Clinical Sciences, Division of Neurology, SE-221 84 Lund Sweden

Background

Stroke is among the most common causes of disability and death worldwide. In ischemic stroke, the stop in perfusion leads to a lack of oxygen and glucose in the affected tissue. In the acute phase, thrombolysis can dissolve the clot and prevent further brain damage. In the subacute phase, much of the damage to the brain is due to the injury of the neurovascular unit that leads to BBB leakage, oedema, inflammation, and then cell death. Currently, there is a lack of stroke treatments to address the subacute phase to halt this process or even restore the neurovascular unit. Being perivascular cells with multipotent and inflammation-modulatory abilities, pericytes could be one of the possible players in these pathogenic processes. We have previously shown¹ that in stroke brain pericytes are activated, migrate from the vessel wall, proliferate and change their morphology, acquiring a microglial morphology and microglial markers expression.

Aim

We are now currently investigating which pro- or anti-inflammatory molecules pericytes express and release after exposure to in vitro OGD (oxygen-glucose deprivation), and the molecular mechanisms and pathways regulating their function.

Methods

Different human pericyte lines are exposed to OGD. Cells RNA is processed with a high-throughput gene expression analysis (Illumina) and compared to normoxic controls' RNA; the secretome of these cells is then analyzed with ELISA on the basis of the gene expression results. With a Western blot approach, the pathway underneath pericytes activation will be finally dissected.

Conclusion

This project will identify previously unknown signaling mechanisms and potential therapeutic molecules that can be exploited for innovation and development of anti-inflammatory therapies in stroke.

- 1 Özen I, et al. (2014) Brain pericytes acquire a microglial phenotype after stroke. *Acta Neuropathol.*

4.5 Francesca Stefani

Anti-tumoral effect of low-dose γ -irradiated

Authors: Stefani FR^{1,2,*}, Eberstål S^{1,2} and Bengzon J^{1,2}

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Glioblastoma multiforme (GBM) is the most common and aggressive primary malignant brain tumor in adults and the prognosis remains very poor despite the current standard of care. Bone marrow-derived mesenchymal stromal cells (MSCs) target glioma metastases efficiently when implanted intratumorally and recent findings demonstrate that MSCs can polarize into immuno-stimulatory cells when exposed to appropriate stimuli. Thus, in the present study we investigated if low-dose γ -irradiation could induce mouse bone marrow-derived MSCs with an immuno-stimulatory phenotype and inhibit brain tumor growth in vivo.

Our results show that following irradiation, MSCs acquire anti-tumoral properties and support the survival of GL261 tumor-bearing mice, with the 5Gy radiation dose being the most effective (29% survival rate). The mechanism is mediated by CD4+ T lymphocytes rather than CD8+ T lymphocytes or NK cells. Further, preliminary results show an increase in the amount of Tbet+ cells within the CD4+ population in the treated group, suggesting a polarization of CD4+ T lymphocytes towards the Th1 lineage. Professional antigen-presenting cells (APCs) may also play an active role in the treatment, as shown by the increased amount of dendritic cells (DCs) in the treated animals.

Besides their stimulatory effect on the immune system, MSCs also affect the immuno-suppressive milieu by decreasing the amount of myeloid-derived suppressor cells (MDSCs) in the blood of treated mice compared to tumor-bearing controls.

In conclusion, low-dose γ -irradiated MSCs represent a simple and fast way for inducing an immuno-stimulatory and anti-tumoral phenotype in MSCs and may represent a favorable alternative approach in cancer therapy.

DATABLITZ - SESSION 5: 11.30 – 12.10, Friday, December 11th, 2015

5.1 Erin Dolan

Characterisation of cognitive dysfunction in an α -synuclein model of Parkinson's disease

Authors: Dolan EK, Nolan YM and Sullivan AM.

Affiliations: Department of Anatomy and Neuroscience, University College Cork

Introduction

Viral vector-mediated-overexpression of α -synuclein in the rodent brain in vivo is a newly-developed model of Parkinson's disease (PD). It reproduces many of the clinical features of PD, including dopaminergic-neuron degeneration in the substantia nigra (SN), decreases in striatal dopamine levels and significant motor impairment. In addition to the characteristic motor symptoms of PD, cognitive dysfunction such as depression, dementia and dysexecutive syndrome can manifest as the disease progresses. Our study aims to investigate and characterise late-stage cognitive dysfunction using the α -synuclein model.

Methods

Adult male Sprague-Dawley rats received into the SN unilateral (3.1×10^8 gc/ 3μ l) or bilateral (two injections of 1.6×10^8 gc/ 3μ l) injection of AAV vector serotype 2/6 overexpressing human wildtype α -synuclein (n=8-10) or GFP (n=6-8). An additional cohort of control animals did not undergo surgery (n=4-8). Animals underwent a series of motor and cognitive tests at various time points throughout the experiment, and were divided into groups sacrificed at either 20 or 40 weeks post-surgery.

Results

Immunohistochemical staining showed widespread transgene expression in the SN at 20 weeks post-surgery, with strong expression in the hippocampus, superior and inferior colliculi and septohippocampal nucleus at 40 weeks post-surgery. α -synuclein-injected animals exhibited significant motor dysfunction in the stepping test, first evident at 20 weeks post-surgery ($p < 0.001$). Olfactory function, measured by an olfactory discrimination task, showed a trend towards significant impairment in the α -synuclein groups ($p = 0.052$). There were no deficits in the conditioned taste aversion protocol in the α -synuclein groups. Interestingly, there were no differences between groups in spontaneous alternations in the Y maze, despite the presence of α -synuclein in the hippocampus.

Conclusions and further work: These results indicate that α -synuclein can accumulate throughout the brain following AAV-mediated-overexpression in the SN, and may play a critical role in PD-related cognitive dysfunction. Further work investigating the protective effects of voluntary exercise on cognitive impairment is on-going.

This work is supported by a grant from Molecular Medicine Ireland. All experiments were conducted in accordance with the European Directive 2010/63/EU, and under an

authorization issued by the Health Products Regulatory Authority Ireland and approved by the Animal Ethics Committee of University College Cork.

5.2 Carol Naughton

Intra-striatal administration of the environmental pesticide rotenone profoundly exacerbates motor dysfunction in the AAV- α -synuclein model of Parkinson's disease

Authors: Carol Naughton^{1,2}, Dan O'Toole³, Eilís Dowd^{1,2}

Affiliations: 1: Pharmacology & Therapeutics; 2: Galway Neuroscience Centre, and School of Medicine, National University of Ireland, Galway.

Although Parkinson's disease is thought to arise as a result of complex interactions between underlying genetics and environmental factors, it is widely modelled in experimental animals using a single genetic or neurotoxic insult. Unfortunately, this has produced models that fail to reliably recapitulate the clinical condition, and this has led to a drive to develop more relevant models with improved validity. One such approach is to combine different risk factors for the disease in order to generate relevant gene-environment interaction models. Therefore, the aim of this study was to assess whether sequential intra-cerebral administration of AAV2/5- α -synuclein and the environmental pesticide rotenone could produce a more relevant model of Parkinson's disease. To do so, male Sprague Dawley rats were assigned to four groups: 1) Control, 2) Rotenone, 3) AAV2/5- α -synuclein and 4) AAV2/5- α -synuclein and rotenone. According to their groups, animals received intra-nigral administration of either AAV2/5- α -synuclein or its corresponding AAV2/5-GFP control, followed 12 weeks later by intra-striatal administration of either rotenone (3.6 μ g) or its corresponding vehicle. Animals were tested at week 8, 10 and 12 after virus administration, and for 5 weeks after rotenone administration on a variety of lateralised tasks to assess motor function. We found that sequential administration of AAV2/5- α -synuclein and rotenone resulted in a profound, stable motor impairment across all behavioural tests employed, and that this was significantly greater than the effect of either challenge alone. These results indicate that the gene-environment approach to modelling Parkinson's disease may produce a more clinically relevant model for testing new and emerging therapies for the treatment of Parkinson's disease.

Acknowledgements: This work was funded through a Postgraduate Scholarship from the Irish Research Council to Carol Naughton.

5.3 Poonam Thakur

Increase in severity of α -synuclein pathology by addition of pre-formed fibrils

Authors: Poonam Thakur*, Ludivine Breger*, Oi Wan Wan*, Kelvin Luk[#], Virginia M Lee[#], John Trojanowski JQ[#], Anders Björklund*

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AAV mediated overexpression of α -synuclein in substantia nigra (SN) has been used to model PD in rodents. However, it lacks spreading of α -synuclein outside the nigrostriatal tract as seen in clinical PD. In our study we added preformed fibrils of human α -synuclein (PFFs) in the rat SN, 4 week after the AAV6-human- α -synuclein (AA6-syn) expression. We observed a significant decline in motor performance of PFF+AAV6-syn injected rats as compared to PFF injected rats. Stepping and cylinder tests indicated that this decline starts to show up around 9 week after PFF injection and continues up to 24 week. Histopathological evaluation at 3 week, 12 week and 24 week post PFF injection indicates that PFF+AAV6-syn causes around 50% loss of TH positive cells in SN as early as 3 week. This loss escalated to around 60% by 24 week. Further, the decrease in TH density in striatum was around 30% by 3 week, which increased up to 60% by end of 24 week. On the other hand, PFFs alone could induce only 20% loss of TH density in striatum in 3 week, which went up to 40% after 24 week. Addition of monomeric α -synuclein instead of PFFs did not cause any accentuation of TH loss. Preliminary analysis indicates that PFF+AAV6-syn cause increase in appearance of phosphorylated α -synuclein, which is indicative of aggregation. This data suggests that ability of external fibrils to cause aggregation of α -synuclein that has been expressed with help of AAV6. Further evaluation will reveal the mode of seeding, transfer (transynaptic versus retrograde) and possible recruitment of endogenous rat α -synuclein. Thus, preliminary studies indicate that addition of PFFs to AA6-syn lead to increase in seeding and toxicity. This model also provides a valuable system to study interaction of human α -synuclein fibrils with wild type α -synuclein in an in vivo environment.

5.4 David J. Harrison

A behavioural characterisation of striatal quinolinic acid lesions in C57BL6/J mice as a model of neural transplantation in Huntington's disease

Authors: David J. Harrison¹, Harry G. Potter¹, Simon P. Brooks¹ and Stephen B. Dunnett¹

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The striatal quinolinic acid lesion (QA) model is commonly used in preclinical cell replacement studies of Huntington's disease, with the success of neural cell grafts in mice often assessed on histological morphology alone. It is vital that the functional viability of grafts is tested to evaluate the effectiveness of the treatment, however a systematic assessment of the deficits in the mouse QA model by which recovery could be judged is required.

This study aims to use an array of motor and cognitive behavioural tasks to establish baseline deficits for which the efficacy of cell replacement therapies can be tested in the QA mouse model of HD.

Two cohorts of C57BL6/J mice were tested on a battery of behavioural tests, including alternation behaviour, memory, attention, anxiety, motivation, activity, skilled reaching, balance, co-ordination, gait and lateral neglect. Thereafter each cohort received QA lesions or sham surgery to either the bilateral dorso-medial (DM) or unilateral dorso-lateral (DL) striatum. Mice were re-tested from one week post-surgery and again from three months post-surgery before perfusion and analysis of the lesions. The position and extent of lesions is to be quantified through histological staining of NeuN+ and DARPP32+ cells.

Preliminary data show that deficits observed are dependent on the position of the lesion within striatum. Mice with DM lesions presented cognitive deficits in the delayed alternation task and a transient increase in footslips on the balance beam but no other motor problems compared to the sham animals. DL lesions however produced deficits on rotarod performance, balance beam, corridor and lateral choice reaction time tasks. Both lesion types induced hyper-activity in open field measures. Some spontaneous recovery was observed by three months post lesioning, limiting the suitability of some tests for long-term transplant assessments. Performance will be compared with the lesion extent following histological analysis.

Acknowledgements: Funded by the MRC and The Wellcome Trust.

5.5 Kiah McCabe

Upregulation of Toll-like receptors in neurotoxic, environmental and inflammatory models of Parkinson's disease

Authors: Kiah McCabe^{1,2}, Ruth M. Concannon^{1,2}, Declan P. McKernan^{1,2}, Eilis Dowd^{1,2}

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The immune-sensing microglial Toll-like receptors (TLRs) are emerging as potential targets to break the cycle of neuroinflammation and neurodegeneration that contributes to the pathology of Parkinson's disease. However, very little is known about TLR expression in animal models of Parkinson's disease which is essential for valid preclinical assessment of the anti-inflammatory and neuroprotective capacity of drugs targeting them. Therefore, the

aim of this study was to characterise the timecourse of TLR expression in Parkinson's disease models induced by neurotoxic, environmental and inflammatory challenges. Male Sprague Dawley rats were given a single intra-striatal injection of 6-OHDA (10 mg), rotenone (1.25 mg), LPS (10 mg) or Poly I:C (20 mg), with corresponding vehicle on the other side, and were sacrificed on Days 1, 4, 14 and 28 post surgery (n=7 per treatment, per time-point). Changes in several inflammatory genes including TLR3, TLR4, microglia, astrocytes and cytokines were examined using qRT-PCR. We found pronounced changes in the bacterial responsive TLR4 and the viral responsive TLR3 receptors in the inflamed striatum in all models, regardless of whether the challenge was neurotoxic, environmental or inflammatory in nature. However, the magnitude and time-course of changes in expression was different between the different models. In addition to the changes in TLR expression, we also found elevations in the microglial marker Cd11b, the astrocyte marker GFAP and both pro-inflammatory (e.g. TNF- α and IL-6) and anti-inflammatory (e.g. IL-10) cytokines in the different models. This study reveals the pattern of changes in TLR expression in different models of Parkinson's disease, thereby highlighting the most relevant model in which to assess the anti-inflammatory and neuroprotective efficacy of TLR drugs. Moreover, it also gives further weight to the suggestion that TLRs may be a useful target for anti-inflammatory disease modification in neurodegenerative diseases where neuroinflammation plays a key role in the neurodegenerative process.

Acknowledgements: This work was funded through a Government of Ireland Postgraduate Scholarship from the Irish Research Council to Kiah McCabe.

5.6 Emma Yhnell

Cognitive training improves disease symptoms in a knock-in mouse model of Huntington's disease

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Huntington's disease (HD) is a rare, incurable neurodegenerative disorder caused by a CAG trinucleotide expansion with the first exon of the huntingtin gene. HD causes a range of motor, psychiatric and cognitive disturbances, for which there is currently no cure. Cognitive dysfunctions are a core feature of HD and are often indicative of a lack of ability to manage independently. Therefore, cognitive training interventions present a potentially exciting non-pharmacological treatment option for neurodegenerative diseases including HD. To test this hypothesis, the HdhQ111 knock-in mouse model of HD was given an intensive session of cognitive training in the 5-choice serial reaction time task (5-CSRTT), an attentional task, for 20 days at 4 months of age. In addition to the group that received

attentional training in the 5-CSRTT, a cage control group received no cognitive training. All animals were then tested in the 5-CSRTT at 12 months of age.

The results suggest that attentional cognitive training, implemented at a young age, improves attentional performance in both wildtype and HdhQ111 animals, at an older age. Attentional training specifically improves motor performance in HdhQ111 animals, as indicated by improved response times in the 5-CSRTT. These results lead to the exciting possibility that specific cognitive training, implemented at a young age, can improve HD disease symptoms later in life.

DATABLITZ - SESSION 6: 14.40 – 15.30, Friday, December 11th, 2015

6.1 Karolina Piracs

Overexpression of mutant huntingtin impairs macroautophagy

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Many neurodegenerative disorders, such as Huntington's disease (HD), are characterized by the formation of protein aggregates in the brain. Genes regulating this protein clearance show an altered expression in HD brains. Better understanding of HD specific alterations in autophagy is crucial for the development of therapeutics. In this project we investigate autophagy in different viral vector based models overexpressing mHTT. To confirm that aggregation of mHTT results in impaired autophagy, we generated lentiviral vectors and transduced 293T cells to overexpress mHTT carrying 66 CAG repeats. We used different autophagy inhibitors to specifically inhibit the different steps of the autophagic pathway. We then performed cellomics scans to measure and compare the amount of LC3 and p62 aggregates in the differently treated mHTT overexpressing cells. We found increased amount of p62 positive aggregates coupled with less LC3 dot number during basal and chloroquine treated conditions. We also performed RNA-seq and found decreased expression for genes involved in cargo recruitment, vesicle trafficking and regulation in mHTT cells compared to the wild type. In parallel, we have generated AAV vectors overexpressing mHTT, performed striatal stereotactic injections in mice and collected samples 10 days, 3 and 8 weeks post-injection. We found significantly more huntingtin aggregates in the cortex and striatum of the mHTT-injected animals, which was not present in the wild-type. We also found striatal fiber loss in the 8 week post-injected animals. We performed western blot analysis on striatal tissue collected from different time points and found different autophagy phenotypes. First we saw an activation of autophagy due to the low number of huntingtin aggregates. 3 weeks after injection we saw the opposite -

autophagy was impaired and by time we observed not only an impairment of autophagy but also an impaired autophagic flux.

Overall our data suggests impairment in later step of autophagy in vitro, but also a more general impairment in vivo.

6.2 Heike Newland

Macroporous microcarriers for neural transplantation and growth factor delivery

Authors: Heike Newland 1*, Petra B. Welzel ¹, Anne Rosser², Carsten Werner ¹, Ben Newland ^{1,2}

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Parkinson's disease is characterized by a specific loss of dopaminergic neurons. Cell therapy is considered a potential means to replace or protect the dying neurons.

Fetal ventral mesencephalic tissue transplantation into the striatum has already shown clinical benefit. However, one big hurdle still needs to be overcome: poor graft survival 1; typically only 5 – 10% of transplanted dopaminergic neurons survive 2. One promising approach is to provide a platform for cells to attach prior transplantation. A lack of attachment is stated as one possible cause of cell death post transplantation in the CNS 3. Motivated by a cell survival increase of 76% reported from a study with poly-L-lysine coated glass beads as implantation platform 4, we developed more suitable injectable macroporous hydrogel microcarriers for cell adhesion and differentiation prior transplantation.

Hydrogel microcarriers were obtained by chemical crosslinking of heparin with four-arm poly(ethylene glycol)₅ in a water-in oil-emulsion. Sub-zero temperature treatment allowed ice crystal formation within the gelling droplets. These ice crystals were removed through lyophilization to leave behind macroporous sponge-like, spherical micrometer-scale structures. These microcarriers have a number of advantageous properties. Firstly, rat mesenchymal stem cells, PC12 cells and fetal ventral mesencephalic cells adhered on them. In comparison to the above mentioned glass beads the microcarriers can also be used for growth factor delivery, which is useful not only for graft survival but also for host neuroprotection. In addition, they use up less dead space in the brain due to the porous structure in which the cells can grow into. Furthermore, the microcarriers with and without cells were injectable through a 30 gauge needle, exhibiting shape memory properties (collapse then reform post injection). Finally, the first in vivo host response data can be presented.

Future studies will analyse whether pre-culturing primary ventral mesencephalic cells to microcarriers improves cell survival in the Parkinsonian rat brain.

(1) Dunnett, S. B.; Rosser, A. E. *Neurobiology of Disease* 2014, 61, 79.

- (2) Björklund, A. *Journal of the Neurological Sciences*, 106, 122.
- (3) Newland, B.; Newland, H.; Werner, C.; Rosser, A.; Wang, W. *Progress in Polymer Science* 2015, 44, 79.
- (4) Jgamadze, D.; Bergen, J.; Stone, D.; Jang, J.-H.; Schaffer, D. V.; Isacoff, E. Y.; Pautot, S. *PloS one* 2012, 7, e30293.
- (5) Newland, B.; Welzel, P. B.; Newland, H.; Renneberg, C.; Kolar, P.; Tsurkan, M.; Rosser, A.; Freudenberg, U.; Werner, C. *Small* 2015, DOI: 10.1002/smll.201500898.

6.3 Gang Wang

Massively Parallel in vivo Screen of Peptide Library Displayed on AAV2 for CNS-directed Gene Therapy

Authors: Gang Wang, Marcus Davidsson, Patrick Aldrin-Kirk, Tomas Björklund

Affiliations: Molecular Neuromodulation, Wallenberg Neuroscience Center, Lund University

Capsid modification is a useful strategy to create adeno-associated virus (AAV) vectors with subtype specific neuronal targeting and enhanced retrograde transport. Incorporating known cell-specific binding ligands is a rational way, but the creation of vectors without a priori knowledge has the potential to reveal novel targets. Directed evolution and phage display are broadly utilized high-throughput methods, but inefficient due to displaying of random peptides wherein the vast majority will be non-functional.

Here, we have developed a novel AAV library in which each virus particle displayed a peptide derived from known neuron-related proteins at AAV2 capsid surface and encoded unique DNA barcode sequences in genome that indicate that peptide. 92,918 unique oligos containing all probable cDNAs encoding 14-amino-acid peptides derived from 135 proteins were synthesized using microarray. Gibson assembly was then used to insert oligos into capsid gene, and barcodes between inverted terminal repeats (ITRs) to generate a plasmid library. Then this plasmid library was used to package a diverse library of AAV virions, such that particles were composed of peptide-modified capsid proteins and genomes encoding barcodes linked to that peptide, which can be used for functional screening by massive sequencing.

Illumina sequencing for the plasmid library showed each peptide could be credibly indicated by unique barcodes. The AAV library was successfully generated and efficiently infected neurons and astrocytes in vitro, which displayed a peptide subset that had efficient retrograde transport ability in neurons in vivo. Functional peptides, which successfully promote neuronal infectivity or retrograde transport, were identified by Illumina sequencing for barcodes in vitro and in vivo. In conclusion, we developed a high-throughput combinatorial method to generate peptide-modified AAV libraries that are valuable for evaluation of receptor expression of neuronal populations and have the potential to generate novel vectors with unique properties for in vivo gene transfer in the CNS.

6.4 Alberto Martínez-Serrano

Electrophysiological properties of human neural stem cells grafted in an in vitro Parkinsonian model

Authors: Alberto Martínez-Serrano¹, Marta P. Pereira¹, Natalia Avaliani², Anna Nelke¹, Merab Kokaia² and Tania Ramos-Moreno^{1,2*}

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Cell replacement therapy in Parkinson's Disease still miss a study addressing the electrophysiological properties of human grafted neural stem cells and their correlation with behavioral recovery after transplantation. Here we study the electrophysiological profiles of C30-Bcl-XL and C32-Bcl-XL, two ventral mesencephalic human Neural Stem Cell clonal lines that express high levels of Bcl-XL to enhance their neurogenic capacity. Electrophysiological recordings show that the majority of the cells derived from the transplants are still maturing at 6 weeks after grafting into the parkinsonian model of coronal striatal slices. One biocytin-labeled recorded C32-Bcl-XL cell showed spontaneous highly regular firing pattern, long duration action potentials, high spike threshold and a hyperpolarization-induced "sag" current. A significant behavioral improvement was observed at 7 weeks post-grafting in the animals transplanted with C30-Bcl-XL, the cell line producing the highest amount of TH+ cells in vitro. A low glial reactivity in the host tissue was also observed.

6.5 Ludivine Breger

Unexpected immune response following the use of retrogradely transported lentiviral vectors in rats

Authors: Breger LS^{a,b}, Lockowandt M^b, Quintino L^b, Isaksson C^b, Lundberg C^b

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Currently, L-DOPA treatment remains the best pharmacological approach to relieve motor symptoms in patients affected by Parkinson's disease. Unfortunately, this medication is associated with disabling side effects, called dyskinesias that develop after few years of treatment. These abnormal, involuntary movements have been shown to correlate with an increase in the ERK (extracellular signal-regulated protein kinase)1 pathway occurring primarily in dopamine receptor 1 expressing medium spiny neurons (D1 neurons)2-4.

Because ERK plays an important role in numerous cell functions, it is paramount to specifically target cells that are affected by the treatment. Specific targeting of D1 neurons is a prerequisite for the development of safe virus mediated gene therapy treatments against dyskinesias. In order to achieve D1 neuronal cells specificity, we took advantage of the FuG-B pseudotype lentiviral vector, which is retrogradely transported⁵. Striatal D1 neurons project in the substantia nigra, consequently, FuG-B lentiviral vectors injected in the substantia nigra will be transported to the cell bodies of D1 neurons in the striatum, thus allowing specific targeting of the direct pathway. Using a reporter gene, we showed that, as expected, injection of the vector in the rat substantia nigra leads to expression of the fluorescent protein in the nigra, as well as the striatum, thanks to retrograde transport of the vector. Better coverage of the striatum was observed 8 weeks after stereotaxic injections, when compared to 4 weeks time points. We showed that it was possible to target striatal medium spiny neurons using MEK-dominant negative FuG-B vector and observed decreasing trend in striatal ERK level. However, the injection of viral vector led to an increased immune response in the substantia nigra and the striatum. We have therefore tested different ways to improve the quality of the virus preparation in order to minimize immune response following FuG-B vector injections.

References:

1. Heiman M et al. 111(12):4578-83, (2014)
2. Darmopil S et al. Biological Psychiatry 66:603-613, (2009).
3. Santini E et al. Journal of Neurochemistry 108:621-633, (2009).
4. Gerfen CR et al. The Journal of Neuroscience 22:5042-5054, (2002).
5. Kato S et al. Human gene therapy 22:197-206, (2011).

6.6 Luis Quintino

Advanced inducible system to regulate neurotrophic factor gene therapy in PD

Authors: Luis Quintino¹, Ludivine Breger¹, Martin Lundblad², Patricia Garcia¹, Christina Isaksson¹ and Cecilia Lundberg¹

Affiliations: 1: CNS Gene Therapy, Department of Experimental Medical Science, Lund University; 2: Developmental Neurobiology, Department of Experimental Medical Science, Lund University,

Glial Cell Line-derived Neurotrophic Factor (GDNF) and associated factors are the only therapeutic proteins able to protect dopaminergic neurons, regenerate axons and improve dopamine release both in animal models and patients suffering from Parkinson's Disease. However, continuous high expression of GDNF in the brain has also significant side effects. To address this issue, we have used a novel inducible system based on Destabilizing

Domains (DD) to regulate GDNF protein expression *in vivo*. We have previously shown that induction of GDNF expression by DD (GDNF-DD) was able to protect neurons in animal models of PD and wanted to validate the system further. After showing therapeutic potential, we wanted to characterize the DD system further.

To evaluate GDNF-DD induction kinetics, viral vectors were delivered to the striatum of animals and expression was turned on over a period of 5 weeks. After expression has reached maximum levels, GDNF-DD expression was progressively turned off. Assessment of biological activity of GDNF-DD used phosphorylation of ribosomal protein S6 (p-RPS6) as a biological activity marker. Maximum biological activity of GDNF-DD was achieved after 3-4 weeks of induction and biological activity of GDNF-DD reverts to basal levels 1 week after system has been turned OFF.

To determine if the effect of GDNF-DD was long-lasting, we repeated the experimental designed used previously. Amperometry assessment of dopamine release in striatum showed a clear 2-3 fold increase in dopamine release of GDNF-DD when compared to controls even after expression of GDNF-DD has been turned OFF for 13 weeks.

6.7 Marina Castro Zalis

Exploration of physical and chemical cues on retinal cell fate

Authors: M.C. Zalis¹, S. Johansson^{1,2}, F. Johansson², U.E. Johansson¹

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Background

Bioscaffolds hold great promise for the advancement of cell-based therapies for retinal neurodegenerations. However, the key physical and chemical factors controlling appropriate donor cell development and subsequent efficient restoration yet need to be elucidated. This can be explored using tailor-made culture assays, where surface nanotopography and presence of extracellular matrix (ECM) proteins and neurotrophic factors can be manipulated.

Aim

We explore whether the substrate nanotopography and presence of ECM protein laminin and neurotrophic factors (BDNF, CNTF) have an effect on the phenotypic differentiation and axonal guidance of retinal cells.

Methods

Mouse retinal post-natal day 4 cells (RPNC) were cultured on electrospun poly-caprolactone (PCL) fiber substrates of random or aligned orientation, with or without laminin coating. Basic (DMEM-F12, B27 supplement) or Full-SATO (Neurobasal, CNTF, BDNF, Forskolin, Insulin) medium was used.

Results

RPNCs could be maintained for at least 18 days on nanosubstrates. At 7 days, increased numbers of retinal ganglion cells (RGC; markers: β -tubulin III, RBPMs, NeuN), photoreceptors (PR; marker: rhodopsin) and glial cells (marker: GFAP) were found compared to 4 h. Addition of laminin in combination with neurotrophins clearly promoted both RGC and PR maturation, demonstrated by complex neuronal morphologies and extensive neurite outgrowth. Nanotopography per se significantly affected formation of cell morphology, with mainly bipolar profiles on aligned fibers and more multipolar profiles on random fibers, independently of the fiber substrate coating or medium used. A remarkable 90° switch of neurite orientation was found after coating with laminin.

Conclusion

We here describe the requirement of both neurotrophins and ECM proteins for extended neurite outgrowth and formation of complex retinal neuronal networks. Most importantly, we for the first time demonstrate that the chemical cue is stronger than the physical cue for the orientation of retinal neurites - a crucial finding for future tissue engineering.

CONGRESS INFORMATION

Previous NECTAR Meetings

Planning meeting	-	1990 Munich, Germany
Founding Meeting	-	1991 Le Vesinet, France
2 nd Annual Meeting	-	1992 Milan, Italy
3 rd Annual Meeting	-	1992 Sandbjerg Manor, Denmark
4 th Annual Meeting	-	1993 Brussels, Belgium
5 th Annual Meeting	-	1994 Amsterdam, The Netherlands
6 th Annual Meeting	-	1995 Maastricht, The Netherlands
7 th Annual Meeting	-	1996 Amsterdam, The Netherlands
8 th Annual Meeting	-	1997 Brussels, Belgium
9 th Annual Meeting	-	1998 Amsterdam, The Netherlands
10 th Annual Meeting	-	1999 Odense, Denmark
11 th Annual Meeting	-	2000 Hannover, Germany
12 th Annual Meeting	-	2001 Brussels, Belgium
13 th Annual Meeting	-	2002 Amsterdam, The Netherlands
14 th Annual Meeting	-	2003 Amsterdam, The Netherlands
15 th Annual Meeting	-	2004 Brussels, Belgium
16 th Annual Meeting	-	2005 Amsterdam, The Netherlands
17 th Annual Meeting	-	2006 Freiburg, Germany
18 th Annual Meeting	-	2007 Lund, Sweden
19 th Annual Meeting	-	2009 Cardiff, Wales
20 th Annual Meeting	-	2010 Freiburg, Germany
21 st Annual Meeting	-	2011 Cambridge, UK
22 nd Annual Meeting	-	2012 Lund, Sweden
23 rd Annual Meeting	-	2013 Cardiff, Wales
24 th Annual Meeting	-	2014 Galway, Ireland

Coming NECTAR Meetings

26 th Annual Meeting	-	December 8th-9th, 2016 Cambridge, UK
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LOST MY CONCENTRATION

ANATOMY WORD SEARCH PUZZLE



ADVENTITIA
CEREBRUM
CLAVICLE
COCHLEA
CRANIUM

CUBITAL
DELTOID
ESOPHAGUS
GALLBLADDER
HUMERUS

METACARPALS
PANCREAS
PATELLA
SCAPULA
SPINAL CORD

STERNUM
TEMPORAL
THYROID
TRACHEA
TRAPEZIUS

