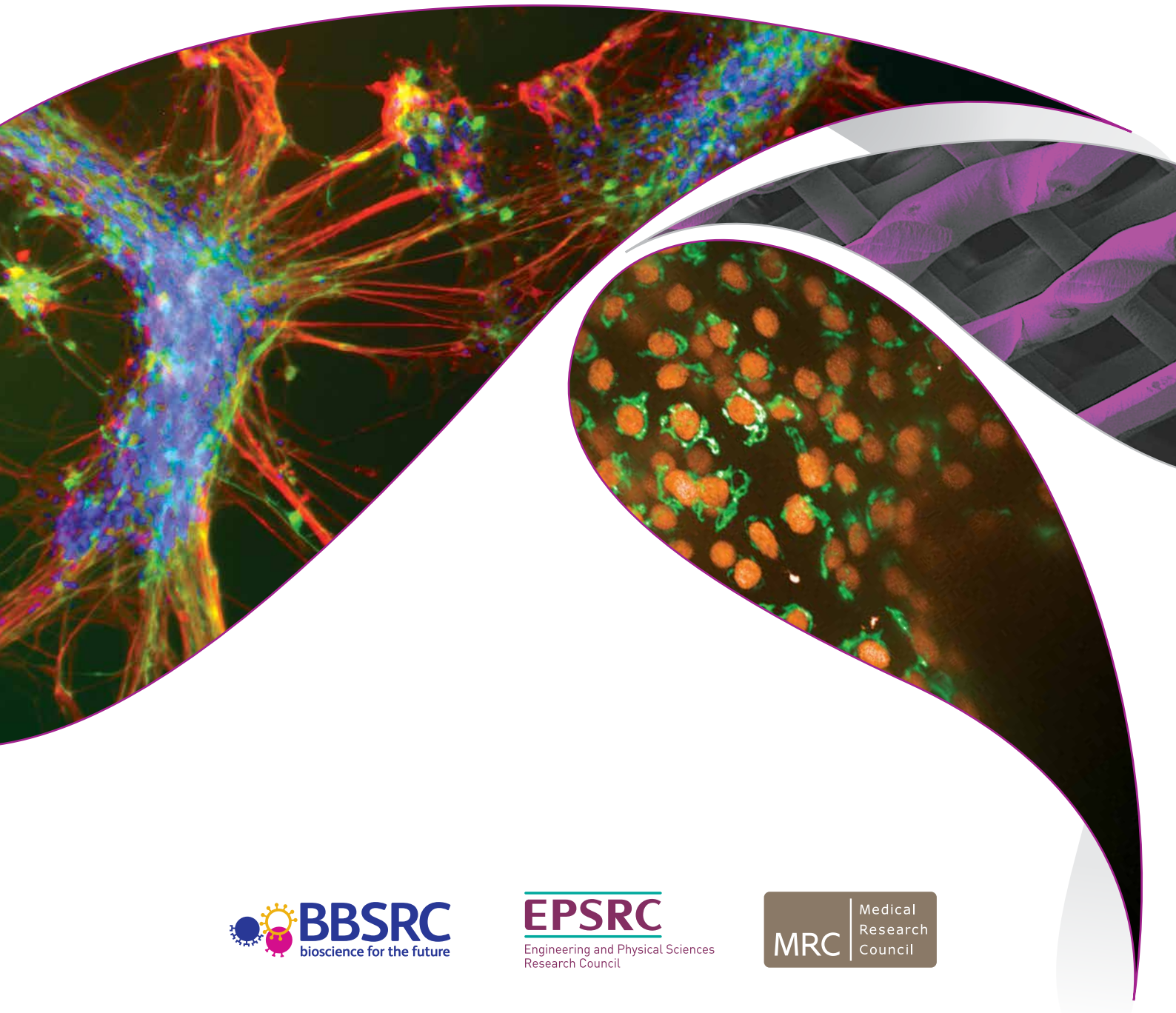
UK Regenerative Medicine Platform

Annual Report 2016



BBSRC bioscience for the future

EPSRC Engineering and Physical Sciences Research Council

MRC Medical Research Council

Cover images

Figs Clockwise Left to Right

Immuno staining of TH (green) and β-tubulin III (red) in differentiated neurons at day 30 derived from the H9 embryonic stem cell lines (PSCP Hub).

3D printed biodegradable scaffolds coated functionalized to bind very low but effective doses of BMP-2. The insert shows the surface of the scaffolds with a fibronectin network and bound BMP-2 (Salmeron-Sanchez Lab).

Expression of albumin (green) and hepatocyte nuclear factor 4a (red) in 3D hepatospheres (Hassan Rashidi; Hay Lab, Niche Hub)

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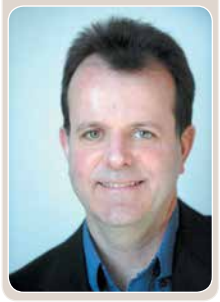


1. Introduction

UKRMP Director: Dr Rob Buckle

The UK Regenerative Medicine Platform (UKRMP) is a national programme seeking to promote the development of regenerative medicine, which holds the promise of revolutionising healthcare over the coming decades through repairing, restoring or replacing damaged or diseased tissues across a wide spectrum of diseases and age-related conditions.

Sponsored by the Biotechnology and Biological Sciences Research Council (BBSRC), Engineering and Physical Science Research Council (EPSRC) and Medical Research Council (MRC), the UKRMP has invested £25M to assemble a cluster of integrated and inter-disciplinary research programmes based upon five central themes. These collectively aim to address the technological and developmental barriers that must be overcome if we are to fully capitalise on the rapid advances being made in the underlying science and translate these towards patient benefit.

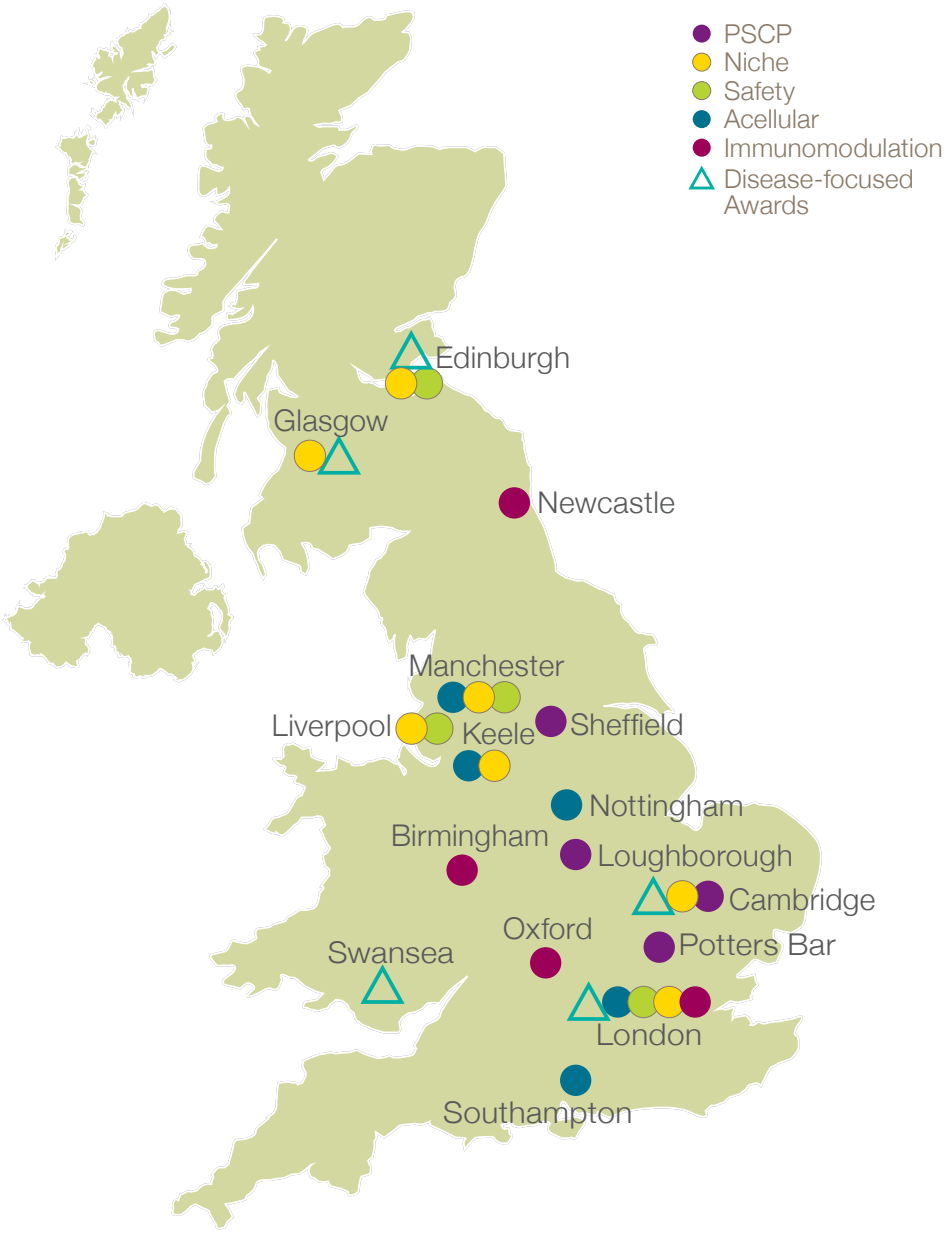
Research within the Platform is focussed on improving our understanding of how the body's regenerative processes can be controlled, manipulated and targeted, and on developing novel tools and technologies to support the testing and manufacture of new regenerative therapies. Since its establishment in 2013, the UKRMP has grown its programme to assemble hubs of activity linking experts in the biological and physical sciences with engineers and clinicians, a network that embraces 17 UK universities. The programme has also developed alliances to support the commercial activity that will be required to bring these emerging therapies to the clinic, working closely with the Cell and Gene Therapy Catapult and engaging with over 25 companies.

The past year has seen the UKRMP's programmes develop a stronger focus on preclinical development, with a number of cross-hub efforts emerging that are coalescing expertise and technologies to address clinical targets such as neural regeneration in Parkinson's disease, liver repair, retinal degeneration and bone and joint repair. Resources and research tools are also becoming available across the UKRMP, in addition to the more routine published outputs of the Platform's research. These encompass research materials such as well characterised cell lines, extracellular matrices, cell scaffolds, and reagents for cell targeting and tracking, as well as access to cutting-edge equipment and the provision of training and support for a wide variety of imaging modalities and manufacturing processes. It is hoped that taken together these will provide a valuable repository through which the research community can readily access validated reagents, research protocols and expert advice, thereby encouraging further cross-disciplinary engagement and collaborative effort.

Importantly, the Platform continues to develop its profile on the international stage, with collaborative programmes now established with groups in France, Germany, Netherlands, Sweden and the USA. These partnerships involve all aspects of the Platform's activity, and span the development, manufacture and preclinical testing of cell therapies, targeted delivery of biomolecules for endogenous repair, and the generation of advanced biomaterials for tissue engineering. UKRMP researchers are also playing a significant role at the global level, where for example they are helping coordinate efforts of leading international groups to ensure shared learning and best practice across themes such as the development of cell therapies for Parkinson's disease and genetic variation and safety in pluripotent stem cells during manufacture and banking.

In the following pages this third annual report of the UKRMP provides further detail of the activities and progress across its five hubs and disease-focu sed programmes. It also highlights some of the interfaces being developed with bioindustry, as well as the emerging outputs from the hub teams that should be of value to the wider community in support of the continued development of regenerative medicine to the benefit of both patients and the UK economy.

UKRMP Hubs And Awards



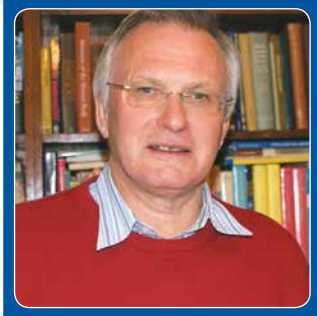
2. Hubs

2.1 Cell behaviour, differentiation and manufacturing Hub
2.2 Engineering and exploiting the stem cell niche Hub
2.3 Safety and efficacy, focussing on imaging technologies Hub
2.4 Acellular approaches for therapeutic delivery Hub
2.5 Immunomodulation Hub

2. Hubs

2.1 Cell behaviour, differentiation and manufacturing Hub

(Pluripotent Stem Cell Platform – PSCP)
Director: Professor Peter Andrews, University of Sheffield



Who

- **University of Sheffield** Peter Andrews, Harry Moore, Marcelo Rivolta and Ivana Barbaric. Zoe Hewitt – Project Manager
- **Wellcome Trust/ MRC Stem Cell Institute, University of Cambridge** Austin Smith, Roger Barker, Ludovic Vallier and Robin Franklin
- **EPSRC Centre for Innovative Manufacturing in Regenerative Medicine, Loughborough University** David Williams, Nicholas Medcalf, Rob Thomas and Mark McCall
- **UK Stem Cell Bank, NIBSC** Glyn Stacey
- **Wellcome Trust Sanger Institute, Cambridge** Mike Stratton and Kosuke Yusa
- **Babraham Institute, Cambridge** Wolf Reik

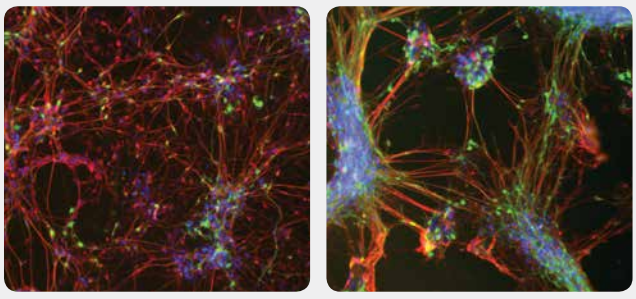
New partners over the past 12 months

- **University of Lund** Malin Parmar
- **University College London** Pete Coffey and Amit Nathwani
- **University of Liverpool** Chris Goldring (Safety Hub)
- Industrial Partners Mathilde Girard (iSTEM, Évry, France) and Heiko Zimmerman (IBMT, Fraunhofer, St Ingbert, Germany)

What

The Pluripotent Stem Cell (PSC) Platform is a translational alliance, combining experts in PSC biology, genetic analysis and clinical cell therapy with leaders in cell manufacturing, safety and regulatory science. We are addressing critical translational bottlenecks by focusing on four key objectives, to:

- Establish protocols for reproducible production, expansion, quality and safety qualification of PSC.
- Develop methods to detect and minimise the occurrence of functionally significant genetic or epigenetic variants during PSC manufacturing
- Standardise PSC differentiation protocols for deriving, manufacturing and banking therapeutically relevant lineage-specific intermediate stem or progenitor cells
- Provide qualified processes for manufacturing regulatory compliant PSC products suitable for clinical use.



Immuno staining of TH (green) and β-tubulin III (red) in differentiated neurons at day 30 derived from the MasterShef7 (L) and H9 (R) human embryonic stem cell lines.

Scientific Developments

PSCP has made good progress against our project objectives over the last year. We continue to address bottlenecks sur rounding translation of potential human PSC-derived therapies including those around safety, stability, quality and manufacturability. One of the areas where this combined effort is having a key impact is in Parkinson's disease.

β

Developing a cell replacement therapy for Parkinson's disease

Parkinson's disease (PD) is a neurodegenerative disorder critically associated with the loss of dopaminergic neurons in an area of the midbrain called the substantia nigra. Humans have around 1 million of these neurons and their death results in an insufficiency of the neurotransmitter dopamine in the regions they innervate, particularly the striatum, an area that controls movement and some cognitive functions deep within the brain. Once 50-60% of these neurons are lost the motor features of the disease, such as tremor, slowness, stiffness and walking/balance problems begin to emerge and steadily progress resulting in major disability and morbidity.

The features of the disease are currently managed by the use of oral formulations of dopamine, but these do not deliver a continuous physiological level of the neurotransmitter and cause complications over time including excessive movements (so called L-dopa induced dyskinesias). An alternative strategy to deliver life-long dopamine replacement in a manner that avoids non-physiological fluctuations is by transplanting replacement neurons generated from pluripotent stem cells to the site where it is most needed in the PD brain.

Research being undertaken in Cambridge by postdoctoral scientist and clinical fellow **Dr Nicholas Blair**, under the supervision of Professor Roger Barker, aims to move cell replacement therapy for Parkinson's disease towards the clinic. This work links into EU FP7 funded (NeuroStem CellRepair) efforts in this field and involves a close collaboration with Professor Malin Parmar and Dr Agnete Kirkeby at Lund University in Sweden. Nick's work has focussed on establishing a robust, clinically suitable differentiation protocol for the generation of dopaminergic precursor cells for transplantation from human embryonic stem cells. He is developing and validating a range of assays for the assessment of cell identity, potency and safety of the final product which will be critical for achieving regulatory approval as the project moves forward towards a first-in-human trial in 2018/19.

This research brings together many aspects of regenerative medicine from within and across the UKRMP Hubs, and collaborative projects are in progress to address some of these including understanding the immune response and the necessary immunomodulation to tolerate allogeneic cells, safety assessments including the genetic stability and biodistribution of transplanted cells, and addressing cell manufacturing and transplantation issues.

Assisting with addressing the cell manufacturing issues, is work being performed by the manufacturing expertise of PSCP, particularly that of **Dr Mark McCall** in Loughborough where PSCP is working alongside the EU FP7 funded NeuroStemCellRepair project to advance this PSC-based therapy for PD.

Process transfer from Lund University into Loughborough University was initiated by a three-person team site visit to the Department for Human Neural Development at Lund University (Sweden). The main aim of this collaboration is to lock down and provide protocols to standardise the current cell-generating process as it moves from development labs to a manufacturing site. Parallel experiments at Loughborough and Cambridge will be used to determine sources of variation and manufacturing risk with the current process so these can be minimised. Mark and the team at Loughborough will also study the stability of both pluripotent and the differentiated cells during cryopreservation. This information will allow the team to decide which methods and technologies can be used to ensure efficient manufacture and functional efficacy of human pluripotent cell-derived neurons to promote functional recovery in PD.

Using this clinical exemplar is providing important opportunities to apply some of the tools developed by the PSCP and wider UKRMP as well as gaining valuable insights into the general process of developing a cell therapy product for clinical use.

Flask preparation for cell culture in a GMP like environment at Loughborough University. This scale of passage process is representative of the scale needed for early clinical manufacture of a dopaminergic neuron product from differentiated pluripotent cells

Hub Growth

Two new co-PIs based at existing partner institutions have joined the PSCP effort. Based at Sheffield University, Dr Ivana Barbaric's research into the molecular basis of aneuploidy in human PSCs and studies of how genetic changes may perturb the normal control of stem cell fate and enhance their ability to grow in culture further adds to our existing expertise. While at Loughborough University, Dr Mark McCall (above), provides expertise in regenerative medicine manufacturing technologies and prediction and reduction of cell therapy cost of goods which also enhances the existing PSCP expertise at Loughborough University.

Partnership Funding

Three new partnership projects have expanded the capabilities and expertise within the PSCP Hub and provided additional opportunities to further interact with other UKRMP Hubs. All three projects compliment the Hub's primary activity to support the translation of PSC cell-based therapies, with a focus on the clinical exemplars of PD and macular degeneration.

As world leaders, recently recognised at the recent 2016 meeting of the International Society for Stem Cell Research, a partnership project between PSCP member Roger Barker with Professor Malin Parmar and her team at Lund University, assists PSCP to contribute and lead on a global platform to the translation of PSC derived Parkinson's therapies-including their work in establishing and developing GFORCE-PD. This whole project aims to resolve one of the bottlenecks to delivering PD cell therapy and that relates to developing robust protocols for the cryopreservation of the final therapeutic cell product and multi-site comparability.

"It is now only 18 years since the first human ES cells were derived by Dr Jamie Thomson in the USA, and less than 10 years since Dr Yamanaka and Dr Thom son first produced human iPS cells, yet clinical trials of PSC-derived cells for several conditions are in progress or on the horizon. In PSCP we are excited by the prospect of contributing to the development of regenerative medicine for Parkinson's Disease."
– Peter Andrews

Calibration verification of a module in the Cedex Bio HT (Roche Life Sciences). The Cedex Bio HT measures a range of cell metabolites and ions from cell culture media to improve understanding of critical process parameters in cell expansion and differentiation

Another bottleneck to progressing PSC-based cell therapies to clinic is one of safety, relating to the genetic stability of this starting material. In a project which combines the efforts of PSCP members Peter Andrews (Sheffield) and Roger Barker (Cambridge) with Pete Coffey (UCL, See Section 3.2) and Chris Goldring (Liverpool) we will be assessing the impact of known and common chromosomal abnormalities that arise in undifferentiated PSCs on the function and safety of therapeutic cell types produced from PSCs for the repair of PD, macular degeneration or liver disease repair.

Industrial collaborations

Cell-therapy contract manufacture and cross site comparability is considered another likely bottleneck in progressing PSC based cell therapies to the clinic. A partnership project which sees PSCP members David Williams (Loughborough) and Glyn Stacey (NIBSC) team up with Mathilde Girard (iSTEM, Évry, France) and Heiko Zimmerman (IBMT, Fraunhofer, St Ingbert, Germany) will look to assess process induced variability across three sites and identify potential controls. To complement this comparability project, PSCP has also leveraged funds from an EU sponsored Project (Scr&Tox, HEALTH-F5-2010-266753) lead by IStem to assist with the development of reference materials for this project.

Finally, the dopamine based programme is working extensively with the Cell and Gene Therapy Catapult to develop a business case for taking the hPSC-derived dopaminergic neuron therapy forward which has also involved seeking further funding for product development and pre-clinical testing.

Networking Activities

PSCP are engaging a broad range of academic scientists, with relevant stakeholders from industry including product manufacturers and developers and their supply chain, clinical users and regulators in a series of workshops.

The first workshop, held in conjunction with the Safety Hub, in Sheffield in January 2015, was a science-based assessment of source materials for cell based medicines. Our second workshop, held at Trinity Hall, Cambridge, September 2015, was on Comparability: Manufacturing, Characterisation and Controls. Meeting reports which capture the perspectives of the attendees on these core issues are now published/in press (See Section 4 and Annex 4).

Two additional workshops are planned to complete the series, one focused on Culture Systems (November 2016), jointly with Plurimes, an EU FP7 project focused on developing mesodermal derivatives of hPSC for applications in regenerative medicine; and a second on Genetic Stability (October 2017). The latter meeting will discuss the issues of genetic stability and its regulatory consequences for regenerative medicine. It will follow a larger international meeting, focused on the underlying biology of genetic variation in hPSC, that PSCP is organising in partnership with the International Stem Cell Initiative (ISCI) in October 2016, at the Jackson Laboratory, Bar Harbor, USA.

In collaboration with the Cell and Gene Therapy Catapult, PSCP has also held in June 2016, a UKRMP wide training workshop on Validation. In collaboration with all five UKRMP Hubs, we held a networking and career development retreat for the UKRMP researchers in March 2016 and we are also organising a joint meeting in conjunction with the British Society for Cell and Gene Therapy to be held in Cardiff in April 2017.

PSCP has also been active internationally; Roger Barker was involved in the organisation of, and contributed to, the ISSCR/ASGCT pre-conference Workshop on Clinical Translation in San Francisco, June 2016. This workshop featured international experts on cellular therapy with experience of moving translational projects into the clinic. Participants gained a broad understanding of the translational process, including early phase trials, and what should be considered for the longer term in taking stem cell based products to market. Finally, Glyn Stacey organised and led the International Stem Cell Banking Initiative Workshop held at CIRM, San Francisco 26th June 2016. This workshop discussed the issues arising from changes in regulation of patient data and quality control standards that are being used by banking facilities for human PSCs.

Future Directions

Moving ahead PSCP aims to continue to focus its research, on supporting the work that is required to clinically translate PSC-based cell therapies for PD and macular degeneration. In particular, in conjunction with other projects and international partners, we will address approaches for both detecting and minimising the appearance of genetic and epigenetic variants during the growth and expansion of PSCs and, crucially, for assessing the potential risks posed by particular variants in different applications

Outputs

- Resources available to the community, see Section 4.
- Publications as a direct result of Hub activities, see Annex 4.

"This collaboration represents a fantastic opportunity to leverage the skills we have continued to develop within the UKRMP program to support an exciting therapy at the early stages of product development where we can have a significant impact. The ability to research the interaction between critical process parameters and a product's critical quality attributes with a clinically destined therapy is an ideal way to demonstrate the methods developed within the UKRMP" - Mark McCall

2.2 Engineering and exploiting the stem cell niche Hub

Director: Professor Stuart Forbes, University of Edinburgh

Who

- **MRC Centre for Regenerative Medicine, University of Edinburgh** Stuart J Forbes, Charles ffrench-Constant, David Hay, Bruno Peault, Anna Williams, Mark Bradley, Pierre Bagnaninchi, James Dear. Jenny Cusiter/Marieke Hoeve – Project Manager, Sarah Neal – Administrator.
- **University of Liverpool** Anthony Hollander
- **University of Cambridge** Robin Franklin, Ludovic Vallier
- **Imperial College London** Molly Stevens
- **Keele University** Alicia El Haj, Ying Yang
- **King's College London** Anil Dhawan, Shukry Habib, Fiona Watt
- **University of Manchester** Sue Kimber
- **University of Strathclyde** Nick Tomkinson

New Partners over the past 12 months

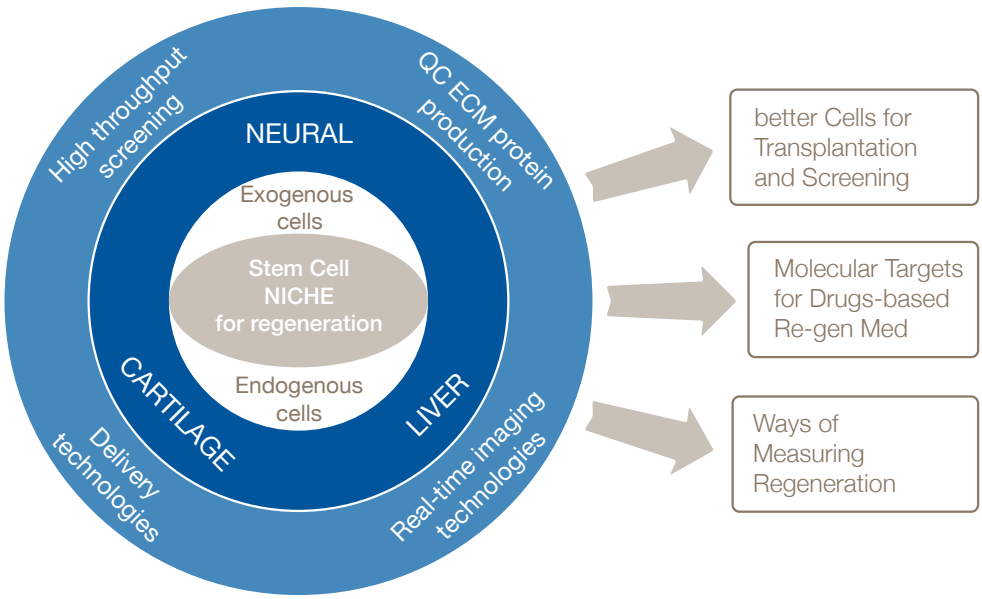
- **King's College London** Tamir Rashid

What

The Niche Hub focuses on understanding and exploiting the signals that stimulate cartilage, liver, and neural tissue repair, to develop tools and technologies for regenerating tissue in man.

We aim to translate the knowledge we accrue from in vitro and in vivo model systems into translational outcomes by taking information from those model systems and applying them to human tissues. The three main approaches our Hub takes to advance regenerative medicine are:

1. Development of better cells for transplantation and screening purposes.
2. Identification of molecular targets for drug-based regenerative medicine.
3. Development of ways to measure tissue regeneration.



Scientific Developments

Highlights of achievements across the Niche Hub for each of our three main approaches are:

Development of better cells for transplantation and screening purposes

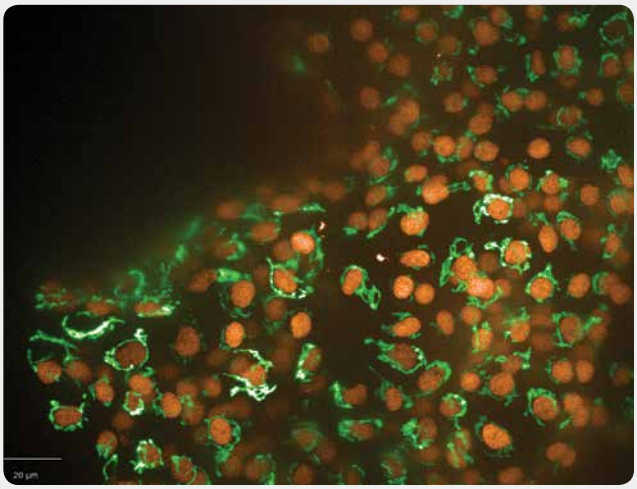
Previous liver research within the Hub has led to the identification and isolation of human Hepatic Progenitor Cells (HPCs) from livers not feasible for transplant. Subsequent work by Wei-Yu Lu, a postdoctoral fellow from the Forbes group has now resulted in the development of two refined methods for isolating HPCs, a cryopreservation method for preserving those cells, and a protocol to culture them in 3D. These outputs contribute to our future goal of transplantation in vivo of HPCs and pluripotent SC-derived hepatocytes.

Our work on identifying synthetic and biological substrates which can support liver stem cell expansion and hepatocyte function has resulted in patented protocols of scalable production of GMP ready hepatocytes leading to improved hepatocyte polarisation, organisation and enhanced metabolic and canalicular function. **Kate Cameron**, a post doctoral fellow from the Hay group, has developed a new technique for growing liver cells from stem cells that is cost-effective and could be adapted for mass production of clinical grade cells. It uses synthetic versions of naturally occurring molecules called laminins as niche support. Alongside improved function and a decrease in unwanted gene expression, these laminins also provide an alternative to animal derived products commonly used. Future work as part of partnership projects aims to encapsulate these cells in alginate based scaffolds for transplantation, in collaboration with Prof Anil Dhawan's team in London.

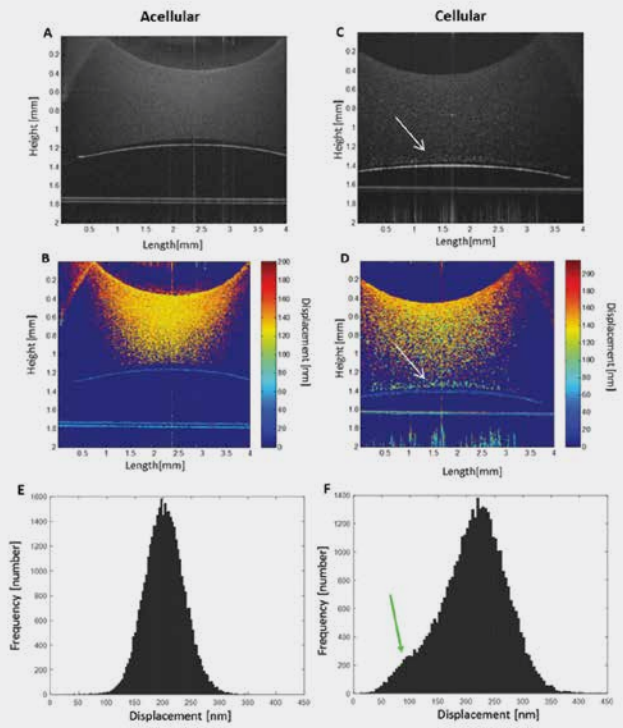
The Hub has developed a new protocol to generate three-dimensional (3D) hepatospheres, which show superior and prolonged metabolic functionality in comparison to 2D-cultured cells. The method is GMP compatible and more cost effective than culturing cells in 2D, and is a promising candidate for translation into cell therapy. To this end, in vivo transplantation of 3D hepatospheres in rodents is currently underway in the Hub.

Protocol development by Stuart Cain, a postdoctoral fellow from the Kimber group, aimed at promoting MSC differentiation towards vascular endothelial cells by modifying key ECM molecules and receptors expressed by MSCs has yielded various molecular tools, including high-yield production of recombinant proteins of superior quality, 3D cell culture models comprising MSC and EC to study correct ECM deposition to increase the structural integrity of vascular grafts for in vivo implantation, and a lentiviral expression system for in vivo live tracking and imaging of fibronectin deposition.

Chondrocytes can be derived from sources such as embryonic stem cells. This offers the potential to use allogeneic cells without invoking the same immune response that mature chondrocytes would trigger. It also allows for a much greater level of expansion than can be achieved with mature chondrocytes. Aixin Cheng and Tao Wang refined a serum-free method to direct hESC through a two week differentiation which utilises exogenous growth factors to mimic the developmental stages experienced in vivo, and validation of the process is currently underway. We have identified several new substrates capable of enhancing pluripotent stem cell-derived chondrogenesis as well as a binding fragment that maintains hESC-derived chondrogenesis. The Hub has shown that a fibronectin fragment including the RGD and syndecan binding domains +/- grapheme oxide can replace plasma or full-length recombinant fibronectin for translation and have established the superiority of BMP-2 over BMP-4 used previously, as part of translation of all reagents to cGMP. Together with collaborators in Manchester, the Niche Hub has brought the pluripotent stem cell-chondroprogenitors to the point of application in large animal joint repair (sheep). Pilot implantation has just been completed, with a longer term trial to be conducted in collaboration with the University of Edinburgh Vet School.



Expression of albumin (green) and hepatocyte nuclear factor 4a (red) in 3D hepatospheres (Hassan Rashidi; Hay Lab).



Structural images of acellular (A) and cell-seeded agarose hydrogels (C) during mechanical stimulation in the hydrostatic force bioreactor (arrow shows cell inclusions). Displacement maps (B, D) obtained by applying elastography algorithms to image data sets. Histograms show displacement distribution for the entire acellular (E) and cellular (F) hydrogels. The arrow indicates a shift in displacement upon cell seeding.

Identification of molecular targets for drug-based regenerative medicine

Work by Eva Borger, a postdoctoral fellow from the Williams group, to promote brain remyelination has yielded several candidate drug targets. These are now being taken forward, including with industrial partners, using mouse models and in vitro cell culture systems. Hub studies have also revealed new insights into myelin sheath regulation, overturning the long-held view that myelin sheath-forming oligodendrocytes are all the same. Oligodendrocytes (a type of glia cell) from different regions were found to generate sheath length on microfibers and neurons that reflect their in vivo origin. These results may help to explain why the response of oligodendrocyte cells to signaling molecules can be quite different between the spinal cord and regions in the brain. This is an important consideration for strategies to repair myelin damage in diseases such as multiple sclerosis, as the oligodendrocytes in the brain and spinal cord might not both respond to treatments in the same way.

Development of ways to measure tissue regeneration.

Mads Bergholt, a postdoctoral fellow from the Stevens Group, has developed novel Raman spectroscopy based techniques for label-free, non-invasive and non-destructive characterisation of cells and tissues. The Hub has developed Raman spectroscopy coupled with multivariate curve resolution (MCR) allowing formation of a 'Raman myelination index', which will be developed further to monitor and quantify the biomolecular compositions of animal and human brain tissues that model Multiple Sclerosis. This will greatly aid our understanding of the progression of this disease and the evaluation of treatments that support remyelination. Mads was recently awarded the UKRMP Special Merit Prize for his activities within the Niche Hub (see Section 6).

The Hub has applied Raman/FTIR spectral analysis coupled with MCR to develop a method for monitoring and quantifying cartilage growth on membranes (e.g. zonal organisation). This revealed the gradients of collagen, GAGs and water in native cartilage and membrane-based tissue constructs, and the methods now being applied for quantitative comparisons of tissue engineered cartilage constructs with native tissue.

The Hub also recently designed a Raman spectroscopic protocol that integrates multiple analytical datasets into one image that can be analysed quantitatively and we are exploiting such advanced techniques to non-invasively monitor liver progenitor differentiation states. Mechano-transduction and cell mechanics are increasingly regarded as important features of stem cell biology.

Yvonne Reinwald, a postdoctoral fellow from the El Haj group, in collaboration with Pierre Bagnaninchi in Edinburgh, has been monitoring the structural maturation of tissue constructs during culture, as their performance prior to implantation into the patient is important for defining their quality, manufacturing criteria and clinical translation.

We have developed a novel optical method which can monitor and measure the 3D constructs online. The new technique links elastography with optical coherence tomography known as optical coherence elastography (OCE) and couples this technique with a hydrostatic pressure bioreactor into a new image modality, HP-OCE. Our results indicate that HP-OCE allows real-time non-invasive monitoring of the displacement and strains of 3D engineered tissue, which enables the investigation of scaffold degradation, material interfaces and heterogeneity as well as changes in scaffold porosity.

Hub Growth

The Hub established five new partnership projects at the end of 2014 which expanded the Hub and its breadth of expertise considerably. All of these projects are progressing well and are delivering tangible outputs. Work on defining a translational niche for tissue engineered products has led to the development of a non-destructive cell imaging platform based on biomechanics, with direct applications in bone and cartilage regeneration research. Research on niche fabrication for chondrocyte differentiation has resulted in the generation of graphene oxide-based substrates capable of supporting hESCs differentiation of mesodermal to chondrogenic progenitors. Work with the Acellular Hub has shown that BMP-2 particles are able to support chondrogenesis to the same extent as soluble BMP-2 and allowed testing of a variety of hydrogels for 3D chondrogenesis. The project aimed at enhancing tissue growth in a dynamic environment shows promising leads for improving bone repair based on findings that implantation of collagen/MSC beads ex vivo in chick femurs leads to enhanced bone regeneration. The role of tethered Wnt has been established in the creation of a platform for directed 3D cues to mesenchymal stem cells in a 3D model using PLGA/collagen. Partnership research to identify new liver toxicity markers has generated a point of care platform for measurement of lead microRNAs in patients with acute liver injury, work which has led to pharma collaboration and a patent.

Networking Activities

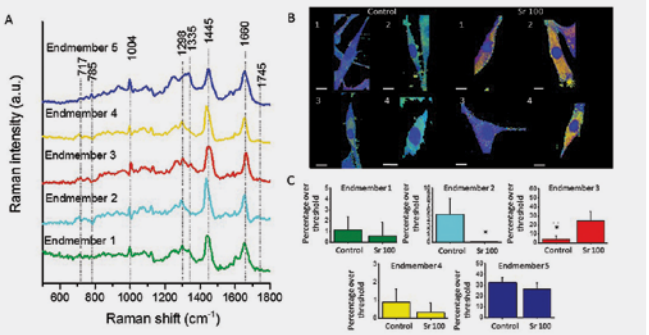
To explore the potential for using Raman Microscopy in regenerative medicine, the Niche Hub, together with OPTIMA-CDT and the Chemistry and Computational Biology of the Niche research facility (CCBN) held a one day workshop in Edinburgh which has resulted in a discussion paper which will appear in print later this year.

The Hub visited the facilities of the National Phenotypic Screening Centre (NPSC) in Dundee to explore high-throughput cell screening. Multiple cell assays are now under development to be run as pilots at the robotics-led screening facility to take advantage of the large compound libraries they have available and accelerate the Hub's small molecule discovery and translational work.

To promote commercialisation of Niche Hub outputs, we hosted visits by pharma companies seeking opportunities to collaborate and expand, and by Scottish Development International and UK Trade & Investment and showcased the variety of tangible outcomes from our Hub that could be of interest for industry collaboration and investments.

Future Directions

The Niche Hub aims to progress regenerative medicine from the laboratory to the clinic through developing direct cell therapies and by targeting and improving the 'endogenous repair' of damaged tissue. To drive our research towards clinical therapies we are keen to collaborate with academia and industry. Our regenerative medicine expertise ranges from material science to bioengineering and cell differentiation, making the Niche Hub an exciting partner for accelerating both academic and industrial R&D and product development activities.



Quantitative multi-image analysis using Raman spectroscopy of cells treated with strontium ions from substituted bioactive glass. (A) Spectra of pure biomolecules identified using spectral unmixing analysis representing proteins (green), various lipids (yellow, red, cyan) and DNA (blue). (B) Colour images representing the relative concentrations of the respective biomolecules. (C) Area based quantification showing the relative abundance of proteins, lipids and DNA in cells. Adapted from Hedegaard, et al. J Biophotonics 2016 9: 542-550.

We are currently successfully engaging with industry in various areas, including 'point of care' platforms, inhibitory molecules, and GMP-compatible cell differentiation methodologies and reagents. We are continuously looking for opportunities to increase this engagement and are keen to discuss ideas and projects with potential new partners.

Outputs

- Resources available to the community, see Section 4.
- Publications as a direct result of Hub activities, see Annex 4.

2.3 Safety and efficacy, focussing on imaging technologies Hub

Director: Professor Kevin Park, University of Liverpool

Who

- **University of Liverpool** Kevin Park, Dan Antoine, Chris Goldring, Neil Kitteringham, Dean Naisbitt (MRC Centre for Drug Safety Science); Dave Adams, Mathias Brust, Marta Garcia-Finana, Raphael Levy, Patricia, Murray, Antonius Plagge, Harish Poptani, Lorenzo Ressel, Matt Rosseinsky and Bettina Wilm. Claire Hutchinson – Project Manager
- **University of Manchester** Stephen Williams, Nick Ashton, Marie-Claude Asselin, Sue Kimber, Kostas Kostarelos, Rachel Lennon and Adrian Woolf.
- **University College London** Mark Lythgoe, Paul Beard, Tammy Kalber, Quentin Pankhurst and Martin Pule
- **MRC Centre for Regenerative Medicine, University of Edinburgh** Stuart Forbes and David Hay
- **University of Glasgow,** Marc Clancy, Patrick Mark, and Rhian Touyz
- **University of Illinois, Chicago,** USA, Natalia Nieto

New partners over the past 12 months

- **University of Sheffield** Peter Andrews (PSCP Hub)
- **University of Cambridge** Roger Barker (PSCP Hub)

What

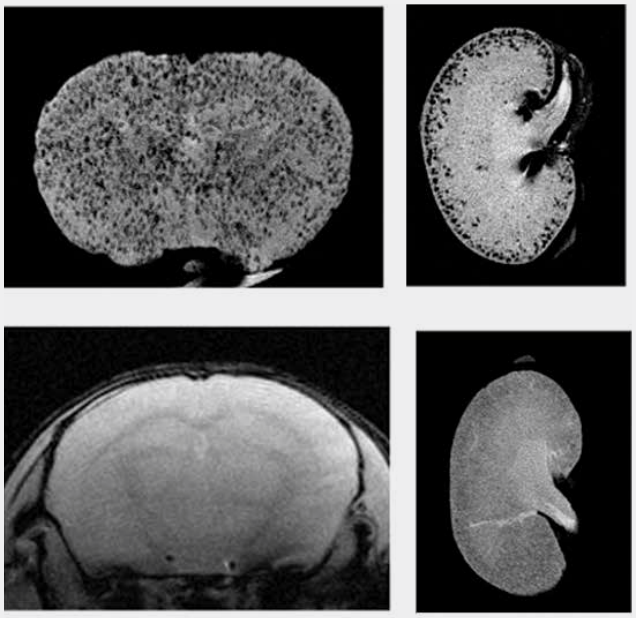
Our focus is to provide a clearer understanding of the potential hazards (and associated risks) of Regenerative Medicine Therapies (RMTs) so that these new medicines can be accelerated into the clinic with full confidence.

We have established a pre-clinical toolkit which gives us the capacity to label any cell type. With flexibility of both chemistry and state-of-the-art multimodal imaging, aligned with mechanistic biomarkers which can be used in assessment of clinical benefit, we can provide a combination that is fit for purpose to address the pre-clinical key issues that determine the safe and efficacious use of RMTs in man. As science is emerging we are in a good position to rollout this multi-faceted toolkit to various disease models.

"To understand the mechanisms of cell therapies from a pharmacological and physiological perspective we need to define the active entity, and the relationship between disposition and effect can inform this." - Kevin Park

Scientific Developments

Over the past 12 months the Hub has employed its novel probes and multimodal imaging technologies, progressing from in vitro assessment to in vivo studies of pre-clinical models of liver and kidney injury to establish which organs the transplanted cells populate and how they affect the function of those organs.



MRI of Super Paramagnetic Iron Oxide Nanoparticles (SPION)-labelled stem cells enables location of the cells to be imaged immediately following delivery. Administration into the left cardiac ventricle leads to whole-body distribution including the brain and kidneys (top). Following intravenous administration no cells (nor SPION debris) are present in the kidney (bottom).

Route of administration for distribution and targeting

Arthur Taylor and Lauren Scarfe working at the Centre for Preclinical Imaging (CPI) University of Liverpool, have shown the route used to administer the cells is crucial as this determines whether the cells reach the target organ and how effective they are.

Using a combination of three imaging modalities, ultrasound for guided injections, bioluminescence imaging for whole body imaging, and magnetic resonance imaging (MRI) for organ-focussed imaging, they have demonstrated that intravenous administration of stem cells leads to the cells being entrapped in the lungs, whereas administration into the left cardiac ventricle enables the cells to be distributed throughout the body. Cells home to the liver when administered intravenously but this is not the case for the kidney and techniques have been developed for targeted administration. Defining the retention and phenotype of cells is of paramount importance for safety.

^{89}Zirconium-oxine celllabelling and magnetic targeting

Stephen Patrick (pictured), working at CABI, UCL is developing non-invasive whole body imaging techniques in the preclinical setting using Positron Emission Tomography (PET) imaging in combination with bioluminescence. By using a combination of imaging modalities and labels, cells can be tracked and their viability assessed over time.

Radiolabelling is the only clinical method that can provide quantitative whole-body biodistribution data; however, the clinical gold-standard, ^{111}Indium oxine, used with Single Photon Emission Computed Tomography (SPECT) imaging can be toxic to cells. The PET tracer ^{89}Zirconium (^{89}Zr) can be used at lower doses than ^{111}Indium oxine due to the enhanced sensitivity and quantification of PET compared to SPECT, thereby reducing toxicity, and offering potential for progression to the clinic. A further advantage of (^{89}Zr) is that it has a relatively long half-life of three days, permitting labelled cells to be tracked for three weeks.

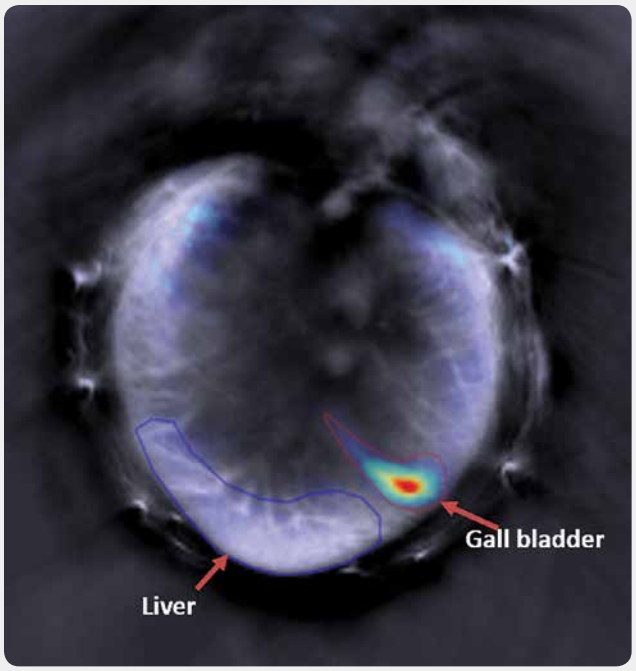
Stephen has also been using the ^{89}Zr PET tracer in combination with Iron Oxide nanoparticles. Delivery and retention are improved by placing a magnetic field at the site of interest towards which passing cells are steered; by activating the magnet the cells can be targeted to a discrete area, in this case the right lung. Through magnetic targeting, therapeutic cells can be more efficiently delivered and retained at the target organ which should enhance the therapeutic effect as well as reduce the number of cells required for injection.

Imaging with Gold Nanorods

Joan Comenge working under the supervision of Raphael Levy, has developed novel Gold Nanorods (GNRs) suitable for cell tracking with photoacoustic imaging using a multispectral optoacoustic tomography (MSOT) scanner.

To demonstrate the potential of GNRs for cell tracking, Joan administered GNR-labelled mesenchymal stem cells into the flank of a mouse; these labelled cells can be easily identified due to their strong photoacoustic signal. It is important to achieve a unique optical signal from thelabelled cells in order to distinguish them from regions with high endogenous absorbance, such as the spleen.

Cells were modified to express luciferase for bioluminescence imaging to validate the in vivo photoacoustic data. This bimodal imaging approach confirmed that the photoacoustic signal originates from the cell clusters and, as the bioluminescence signal can only be generated by living cells, the GNRs were not affecting cell viability. Furthermore, a time course analysis indicated GNRs did not affect the proliferation or differentiation of the



MSOT image of ICG fluorescent dyes to asses clearance through the liver and gall bladder of a mouse. Clearance is delayed in injury models compared to control which correlates with biomarker and histology data.

cells. Working with Jack Sharkey, Joan has also labelled macrophages with GNRs, observing homing to the liver following systemic administration.

Following successful detection and viability assays, GNRs are now available for use across the Safety Hub. Collaborator Tammy Kalber (UCL) has analysed these novel probes in their unique photoacoustic system which is complementary to MSOT as it allows greater sensitivity to very few cells at very high spatial resolution. The GNRs are now being used in combination with other tracking agents and imaging modalities as a tool to precisely monitor cell engraftment in animal models.

Emphasis on functional imaging linked to biomarkers

We can translate our pre-clinical findings using complementary methodologies across the Hub by combining biodistribution studies with functional imaging, mechanistic biomarkers and histopathology. This triangulation approach enables us to quantify the efficacy of therapies and produce a safety platform, with broad applicability, to show enhanced function, amelioration of tissue damage and regeneration with read across to man.

Hub Growth

The Hub has increased links with the Pluripotent Stem Cell Platform (PSCP) Hub and this year will be commencing two projects utilising our expertise and novel technologies to answer specific scientific questions relating to biodistribution of cells in a Parkinson's Disease model, and assessment of the tumourgenic potential of a known, frequent human embryonic stem cell genetic variant in a liver engraftment model. The latter project in particular is a multi-Hub collaboration which involving the expertise from members across the Safety, PSCP and Niche Hubs.

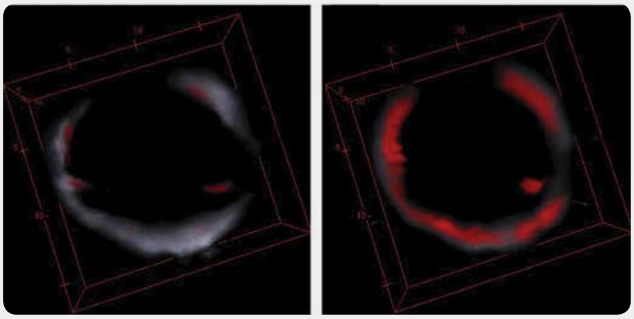
Networking Activities

Following the Hub's Nanoparticles for Cell Tracking work shop last year, further collaborations have developed and the meeting led to Safety Hub involvement in a complementary global programme coordinated by Health and Environmental Sciences Institute (HESI) on the emerging issue of Cell Therapy – Tracking, Circulation and Safety; its mission is to establish a platform where an international network of experts from multiple sectors share knowledge in the rapidly evolving field of cell therapy.

The Safety Hub is planning two further workshops for 2016; Pre-clinical Imaging for Safety Assessment in conjunction with the MRC Centre for Drug Safety Science and a joint meeting with the UKRMP Immunomodulation Hub exploring the activity of MSCs for scientific and clinical use.

Future Directions

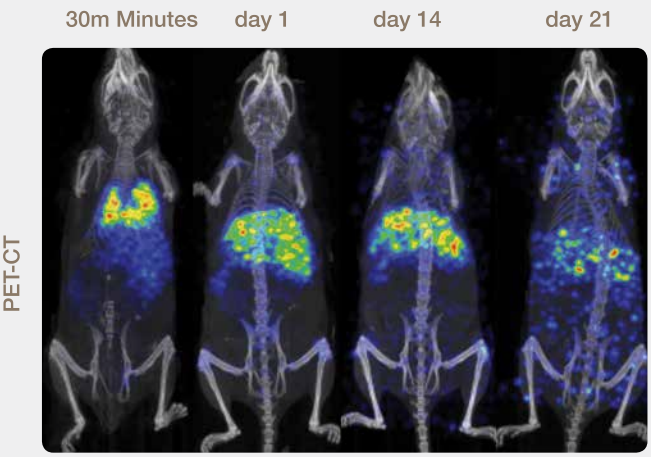
Building on our expertise we have developed a roadmap for translation of cell therapies which will align all the techniques developed during the current programme and demonstrates the Hub's trajectory towards clinical delivery of regenerative medicines. By assessing the right label (for tracking and function) and using the right imaging modalities we can assess safety through biodistribution in healthy animals before progressing to animal models of disease, with relevance to particular human applications, for mode of action and safety and efficacy assessment linked to clinical need.



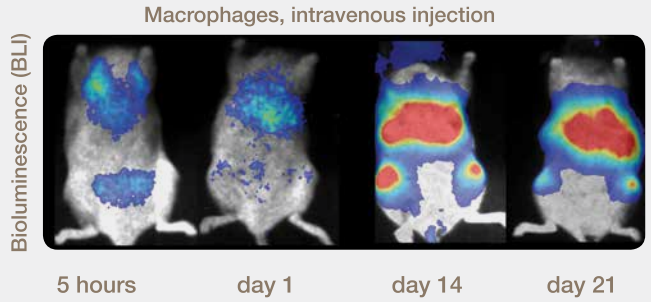
Accumulation of GNR-labelled macrophages in the liver over time using MSOT following systemic administration. The red colour (right) indicates the regions that match the spectra of GNRs six hours after administration.

Outputs

- Resources available to the community, see Section 4.
- Publications as a direct result of Hub activities, see Annex 4.



Macrophages expressing firefly luciferase can be labelled with a low dose of 89Zr-oxine and then tracked with PET and bioluminescence for 3 weeks (panel above). Bioluminescence images show cells are viable and confirm their location (panel below).



"Through our strengths in multimodal imaging and chemistry we have developed a toolkit that is fit for purpose across a wide range of pre-clinical disease models. In combination with mechanistic biomarkers and histology this provides a platform for translation to man." - Kevin Park

2.4 Acellular approaches for therapeutic delivery Hub

Director: Professor Kevin Shakesheff (pictured), University of Nottingham
Co-Director: Professor Molly Stevens, Imperial College London

Who

- **University of Nottingham** Kevin Shakesheff, Felicity Rose and James Dixon. Sharon Crouch – Project Manager
- **Imperial College London** Molly Stevens
- **University of Southampton** Richard Oreffo
- **Keele University** Alicia El Haj
- **University of Manchester** Julie Gough, Sue Kimber (Niche Hub), Ailine Miller and Stephen Richardson
- **Cardiff University** Alastair Sloan
- **University of Birmingham,** Liam Grover
- **MRC Centre for Regenerative Medicine, University of Edinburgh** Stuart Forbes (Niche Hub)
- **University College London** Robin Ali, Richard Day
- **University of Cambridge** Stephano Pluchino
- **University of Liverpool** Sajjad Ahmad, Rachel Williams
- **Clinical Spokes include** James Fawcett (Cambridge), Philip Newsome (Birmingham), Sheila MacNeil (Sheffield), Ilyas Khan (Swansea), and Krish Raganuth (Nottingham

New partners over the past 12 months

- **University of Liverpool** Rachel Oldershaw and Thomas Maddox
- **Aintree University Hospital,** Michael McNicholas
- **Newcastle Surgical training Centre,** David Deehan
- **University of Paris Descartes,** Philippe Menasché
- **King's College London,** Fiona Watt (Immunomodulation Hub)

What

This Hub has adopted a multi-disciplinary approach in the design of a wide range of materials, both from synthetic and natural sources to enable:

- Cell survival and function at the intended site of regeneration
- Localisation of drugs to augment tissue regeneration
- Guided tissue self-assembly in 3D architectures

Our key objectives relate to new technologies that enhance the efficacy and safety of future regenerative medicine products to create an environment in vivo that facilitates tissue formation. Therapeutic delivery systems build on the principles of biomaterials design and drug delivery to create final products in which the efficacy of cell therapies or the mobilisation of the patient's own stem cells are maximised.

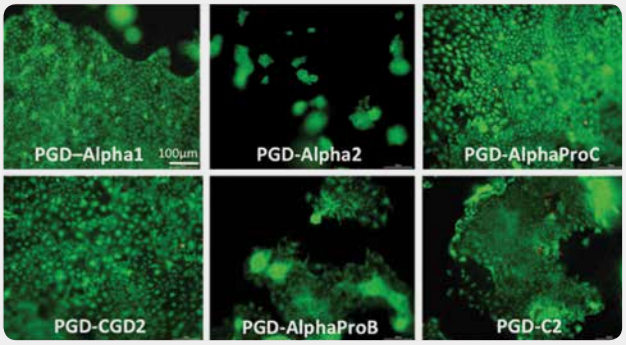
In this our third year of technology development, significant advances continue to be made particularly in the area of intra cellular delivery, the use of biomaterials to prevent fibrosis and successful pre-clinical investigations in a number of disease areas.

Scientific Developments

Injectable Hydrogel for the treatment of Barrett's Oesophagus

The drawbacks of current management strategies in reducing post-operative strictures in patients with Barrett's Oesophagus has led to novel research developing alternative strategies to better prevent strictures from forming.

Dr Deepak Kumar (University of Manchester; has been working closely with the clinical team in Nottingham, led by Dr Krish Ragnuth, to develop a ready-to-use, synthetic, self-assembling peptide hydrogel that can be administered endoscopically to prevent post-operative scar formation. Novel peptide hydrogels have been supplied through the collaboration with Professor



Viability of mouse oesophageal epithelial cells (mOECs) after 3 days of culture on top of a panel of buffered peptide hydrogels.

Alberto Saiaini (University of Manchester); these have been tested against stromal fibroblasts and epithelial cells for their potential to support cell survival and expansion; injectable/sprayable formulations are now being developed. Julie Gough's group have now established an international collaboration with Dr Steve Badylak's team in Pittsburgh to evaluate the muco-adhesiveness of peptide hydrogels to normal and Barrett's oesophageal tissue in addition to using their extracellular matrix based material as an inductive template for tissue remodelling.

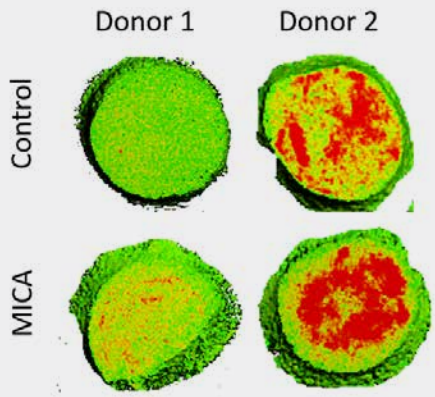
Protein Fusion Technology

Protein transduction domains (PTDs) are powerful non-genetic tools that allow intracellular delivery of molecules to modify cell behaviour. This technology has been developed by Dr James Dixon (University of Nottingham) to provide a strategy for controlling cell labelling and directing cell fate or behaviour that has broad applicability for basic research, disease modelling, and numerous clinical applications. A patent has filed on this work and is published in PNAS (see Annex 4).

The delivery system is now being used by a number of the Hub partners to program human mesenchymal stem cells into bone and cartilage for orthopaedic regenerative medicine. In particular **Dr Robin Rumney** (University of Southampton) has been investigating enhanced intracellular delivery of factors to accelerate angiogenesis and osteogenesis. Ultimately this work will be translated to delivery of these protein therapies for promotion of intervertebral and long non-union bone healing.

Magnetic nanoparticles

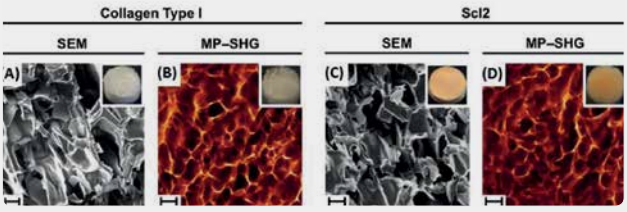
One particular challenge for bone re-growth lies in the delivery of functional mechanical stimuli to implanted cell populations to activate and promote osteogenic activities. **Dr Hareklea Markides** (Keele University) has been developing novel bio-magnetic approaches to overcoming this challenge. Magnetic mechano-activation defines a novel biomagnetic approach to remotely deliver mechanical stimuli directly to individual cells. The group at Keele, together with Dr Jane McLaren at the University of Nottingham have developed a magnetic nanoparticle based approach to promote bone growth. Ultimately this technology will be used in the treatment of non-union bone fractures and skeletal defects.



µCT images of donor responses to MICA magnetic activation. TREK-labelled MSCs encapsulated within a collagen hydrogel (2.5mg/ml) and magnetically stimulated (1hr/day over 28 days) increased mineralisation (red) across donors compared to non-stimulated counterparts.

Polymer-peptide hybrid scaffolds

Targeting epithelial-to-mesenchymal transition (EMT) as a strategy to prevent fibrosis is gaining momentum. Thus far, efforts have mainly focused on the systemic administration of drugs that block pathways involved in EMT. Clinical trials of these therapeutics have highlighted secondary and off-target effects. Our strategy addresses the limitations of these approaches by preventing EMT in a localised manner. Drs Jenny Puetzer and Jean-Philippe St-Pierre, based in Prof Molly Stevens' lab at Imperial College, have functionalised electrospun PCL membranes to act as anti-fibrotic coatings in acellular implants. Work is currently on going in pre-clinical mouse models of peritoneal fibrosis. The Stevens labs have also output scaffolds based on Streptococcal collagen–like 2 proteins that inherently lack cell–binding sites and thereby provide a structurally well–defined biological 'blank slate' by which to systematically integrate specific motifs for a desired cellular response, as guided by the inclusion of hyaluronic acid (HA)-binding and chondroitin sulphate (CS)-binding peptides.



SEM images of C) collagen type I and E) Scl2 foams (scale bars are 100 µm). Representative MP-SHG images of D) collagen type I and F) Scl2 foams (scale bars are 200 µm). Insets show photographs of collagen foams of 8 mm in diameter. Adapted from: P.A. Parmar, et al., Advanced Healthcare Materials. 2016: 10.1002/adhm.201600136.

Hub-to-Hub Collaborations:

One focus for the Niche Hub is influencing cell differentiation in cartilage, neural and liver repair. Dr Omar Qutachi (University of Nottingham) has been working with the Niche Hub to provide biodegradable polymer particles for the enhancement of cell engraftment in liver (Stuart Forbes, Edinburgh) and to promote chondrogenic differentiation for cartilage repair (Sue Kimber, Manchester). These particles have been developed to facilitate the slow release of specific growth factors at precise locations within the body. The approach is transferable across many drug and tissue types and is a platform technology that the Hub is keen to provide to new collaborators.

Working with collaborators Dr Raul Elguata, and Professors Giovanni Lombardi and Prof. Fiona Watt of the Immuno-modulation Hub, **Dr Derfogail Delcassian** (pictured) is investigating the use of acellular materials to direct

regulatory T cell behaviour in skin graft therapies. This project combines expertise in materials based immunomodulation from Nottingham, with partial skin graft transplantation expertise at King's. Using T cell stimulatory biomaterials, this project aims to selectively recruit and expand populations of T regulatory cells to the transplant site, whilst suppressing the activity of other T cell phenotypes which lead to graft rejection.

Hub Growth

The last year has seen a significant expansion of Hub activities with international partners. A number of new projects have commenced this year bringing with them global expertise in key areas. The previously mentioned collaboration between Manchester and Steve Badylak's group in Pittsburgh is one of 4 exciting new international Hub collaborations.

Dr Rachel Oldershaw (University of Liverpool), Mr Michael McNicholas (Aintree University Hospital), Prof David Deehan (Newcastle Surgical training Centre) and Dr Thomas Maddox are working with the Hub on the development of a medical device to support the delivery of cell therapies in surgery. With ruptured anterior cruciate ligaments (ACLs) there is a need to enhance the integration of the graft into the bone to allow for effective restoration of knee function and to reduce the incidence of re-rupture. The group are working with the Chicago based company BioPoly®.

Molly Stevens' Group at Imperial are collaborating with Professor Philippe Menasché, University of Paris Descartes, to develop biomaterial-based approaches to deliver extra cellular vesicles/exosomes (EVs) for cardiac tissue repair. With heart failure affecting 6-10% of people over the age of 65 years worldwide and resulting in the deaths of approximately 30-40% of patients within a year of diagnosis, cardiac tissue repair needs to be addressed by the most state-of-the-art approaches. It has become increasingly evident that cell therapy had to be combined with some form of tissue engineering strategy; this collaboration with the use of EVs could improve the materials-based delivery approach. Stevens' research assessment has now performed some initial in vivo rodent experiments in the Menasché Group.

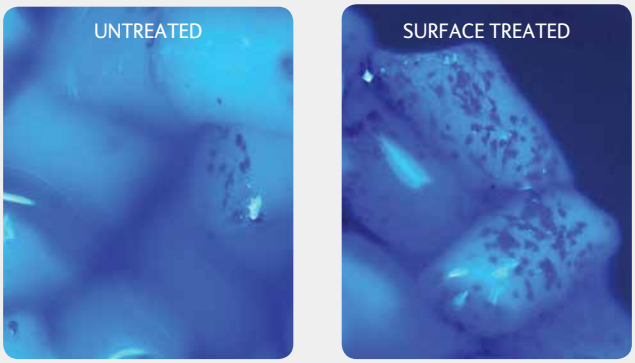
In collaboration with Prof. Anderson and Prof. Langer's labs at MIT, Derfogail is developing new materials for islet trans plantation in Diabetes-1. These therapies use selectively

functionalised controlled-release microparticles, containing small molecule immunomodulators that can direct innate and adaptive immune cell behaviour. Using these materials, in conjunction with chemically modified hydrogels, Derfogail can encapsulate islet cells to provide a 3D transplant scaffold that supports islet cell function and simultaneously modulates the host immune response at the in vivo transplant site. These materials will allow for greater control of the host immune system behaviour in the transplant niche, helping to control fibrosis and increasing transplant graft survival, and are applicable to many cell transplant therapies within tissue engineering and regenerative medicine (TERM).

Networking Activities

Identifying the commercial potential of all Hub technologies and scoping out the route map to clinic was the focus of a Commercialisation Meeting held at Imperial College in January 2016. Acellular Hub academics 'pitched' their technologies to a panel of industry experts and that feedback is now being implemented in the planned development of a number of technologies.

The Acellular Hub members have enjoyed a large number of keynote/planary invitations including TERMIS World Congress (Boston), Bone-Tec International Congress (Stuttgart), Engineering Life (Dresden) and the Southampton Science Park's Inaugural Christmas Lecture. Kevin Shakesheff and Felicity Rose took part in the 2016 Cheltenham Science Festival, presenting on general concepts in tissue engineering and demonstrating cutting edge 3D bioprinting via a live feed to Nottingham. Richard Oreffo and the Bone and Joint Research Team have taken their unique Stem Cell Mountain exhibit and presented at EU_TERMIS (Uppsala), Glastonbury and various schools/ festivals to enhance public engagement and understanding of regenerative medicine. Kevin Shakesheff now sits on the Restoration of Appearance and Function Trust's (RAFT)



Effect of surface modification on the attachment efficiency of human mesenchymal stem cells (toluidine blue labelled) to TAOS™ injectable tissue scaffold pellets

Scientific Advisory Committee; the charity's aims and re search focus in bone regeneration and wound healing aligns well with the Acellular Hub and they will be a key networking partner going forward.A main focus of the Hub in the remaining months will be to expand networking activities, meetings with industry, key opinion leaders and regulators.

Future Directions

The last 12 months has seen a shift from the development of Hub core technologies to further development in the pre-clinical models. Key areas of focus over the remaining 12 months will be on bone repair, anterior cruciate ligament reconstruction and completion of pre-clinical studies on enhancement of liver and neural engraftment. We are now at a stage where potential hurdles to clinical adoption are being addressed, these include end user needs, regulatory and manufacturing and scale up.

Industry collaboration is a major focus for the Acellular Hub and Dr Felicity Rose (University of Nottingham) is the academic PI on an Innovate UK project with Locate Therapeutics to optimise self-assembly scaffolds for delivery of cells to sites of tissue regeneration. Migration of cells away from the required site of action has posed a significant problem for cell based therapies. Nottingham are modulating the cell surface chemistry of the scaffold particles to allow attachment and delivery of cells; these will then be tested by Locate in a series of pre-clinical models for scaffold performance and retention of cells at the site of administration.

The Hub remains keen to work with new groups who wish to improve cell and drug delivery and welcome external approaches by international groups and companies to engage in collaborative translational research.

Outputs

- Resources available to the community, see Section 4.
- Publications as a direct result of Hub activities, See Annex 4.

2.5 Immunomodulation Hub

Director: Professor Fiona Watt, King's College London

Who

- **King's College London**, Fiona Watt, Francesco Dazzi; and from the MRC Centre for Transplantation, Giovanna Lombardi and Steven Sacks
- **University College London** Robin Ali
- **The Francis Crick Institute** Caetano Reis e Sousa
- **University of Oxford** Paul Fairchild and Fiona Powrie
- **University of Birmingham** Philip Newsome
- **Newcastle University** James Shaw
- **Imperial College London** Sian Harding

New partners over the past 12 months

- **Imperial College London** Marcus Dorner
- **University of Cambridge** Stefano Pluchino (Acellular Hub)
- **University of Nottingham** Kevin Shakesheff (Acellular Hub)

What

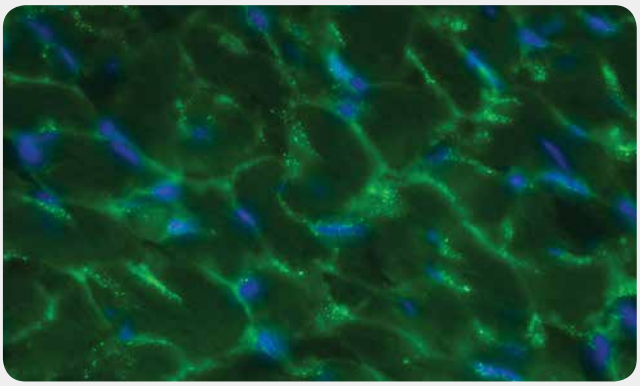
We are pooling our collective knowledge and sharing experimental tools to answer three questions:

1. How do differentiated cells signal to the host innate and adaptive immune system?
2. How do transplanted cells provoke adaptive immune responses?
3. How does the inflammatory niche contribute to endogenous repair and influence the fate of transplanted cells?

Scientific Developments

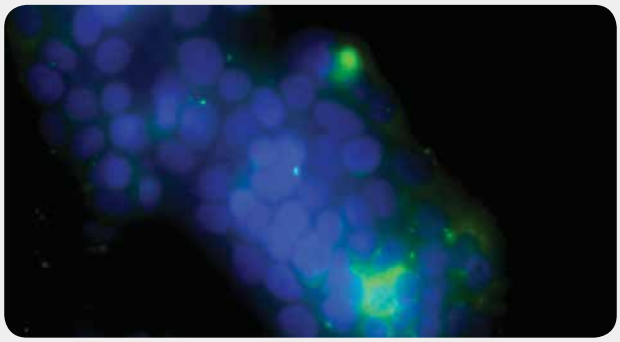
How do differentiated cells signal to the host innate and adaptive immune system?

There is particular interest in the therapeutic potential of cells that have been differentiated from pluripotent stem cells, but their immunogenicity is poorly understood. Our focus here is to carry out a systematic analysis of how differentiated cells signal to the immune system. We are achieving this by comparing human cells differentiated from induced pluripotent stem cells with cells isolated directly from the corresponding adult tissue. Particular tissues of interest for the Hub are hepatocytes and retinal pigment epithelium (RPE). Adult hepatocytes are being provided by our investigators in Birmingham (Philip Newsome and his research associate Jasmine Penny) and Niche Hub collaborator Anil Dhawan. iPSC-derived hepatocytes are being provided by Tamir Rashid and David Hay (Stem Cell Niche Hub), while Robin Ali and his research associate Pete Gardner have been generating iPSC-derived RPE. A third tissue type is also being evaluated, iPSC-derived cardiomyocytes, which are being provided by Sian Harding from Imperial.



Immune-complexes of Immunoglobulin G (green) deposited in the myocardial cells from a mouse with immune-mediated heart tissue damage. Nuclei are labelled blue.

The immunogenic properties of iPSC-derived cells are dynamic and largely dependent upon their exposure to stress, which increases during transplantation Our investigators at King's College London (KCL, Giovanna Lombardi, Steven Sacks and their research associates Raul Elgueta and Giorgia Fanelli) have completed immune profiling studies of iPSC-derived hepatocytes and RPE cells under both steady state conditions and when exposed to IFN-γ treatment or to hypoxia, a process that leads to activation of pro-inflammatory and fibrotic pathways in susceptible cells. Fang Xiao from Giovanna Lombardi's laboratory has started the immune profiling of iPSC-derived cardiomycytes under the same conditions described above. They have been able to quantify the expression of CD80, CD86, MHC-II (HLA-DR), PDL-1 and CD40 in these cells. They have established that none of the cell types express co-stimulatory molecules, and while the iPSC-derived RPE cells express HLA-DR following IFN-g treatment, iPSC-derived hepatocytes do not.



HepG2 hepatocytes (blue) subjected to oxidative stress showing C3C complement activation (green).

In addition, they have analysed the cytokines produced by the different types of iPSC-derived cells under the conditions described above. Raul, Giorgia and more recently Fang have also been examining the effects of iP SC-derived cell types on allogenic T cell proliferation and cytokine production. They have also found that iPSC-derived hepatocytes and RPE cells do not induce T cell proliferation in vitro and that stimulation of T cells by anti-CD3/CD28 beads or by allogeneic dendritic cells is inhibited in the presence of the same cells.

These findings are being used to compile response signatures that will act as predictors for the likelihood of an immunogenic response from iPSC-derived cells before they are selected for transplantation. Our studies have identified Collectin-11, a soluble C-type lectin that activates the lectin pathway of the complement system and the complement component C3d as a key component of the immunogenic response of transplanted cells. Specifically, Giorgia has determined that Collectin-11 is unregulated in iPSC-derived RPE cells under hypoxic stress conditions and that this causes an immunogenic response in transplanted cells. Giorgia is currently working on ways to attenuate the upregulation of Collectin-11 in transplanted iPSC-derived RPE and these studies will be extended to other cell types.

How do transplanted cells provoke adaptive immune responses?

This year, we have progressed from in vitro studies to analysing cell behaviour following transplantation. Pete Gardner and **Giorgia Fanelli** are going to transplant iPSC-derived RPE into humanised mouse models (NOD/SCIDγc-/--NSG mice reconstituted with either peripheral blood mononu clear cells (PBMCs) or with human CD34+ cells) in order to evaluate the different pathways of allorecognition in vivo. These studies will inform on the conditions required for future studies in the Hub, which are aimed towards achieving successful transplantation and sustained engraftment of iPSC-derived RPE in mouse models with blindness-inducing retinal tissue damage.

Research associate Fang Xiao is a new addition to the Hub this year who is focusing on transplantation for liver repair. This work is based on her previous findings which showed that pre-treating human islets with a membrane-localizing complemet C3 convertase inhibitor, attenuates early allograft damage that occurs after transplantation underneath the kidney capsule in humanised mice (NSG mice reconstituted with CD34 cells). In addition, she has also previously shown that the rejection of human adult islet cells transplanted under the kidney capsule in the same mice is delayed when the mice are injected with human regulatory T cells (Tregs). Using her expertise with this model, Fang has started to evaluate the optimal number of iPSC-derived hepatocytes that can be transplanted under the kidney capsule of NSG mice in order to study the immune response after reconstitution by measuring the release of albumin. Following on from Giovanna Lombardi's previous studies - if an immune response is detected against iPSC-derived hepatocytes - Fang and Raul will be investigating the effects of either of the two treatments (C3 convertase inhibitor or Tregs) in isolation first and then in combination on sustaining engraftment of iPSC-derived hepatocytes. Furthermore, Fang and Raul are collaborating with Marcus Dorner at Imperial College to compare the behaviour of iPSC-derived and adult hepatocytes following transplantation into a new humanised

liver injury model that has been established by Marcus. In this model the damaged liver of humanised mice is repaired by injecting hepatocytes directly in the spleen. This model will also be used to assess the effectiveness of the combined Treg-complement inhibitor treatment in facilitating repair of liver injury. Once fully established, this new mouse injury model will be made available to other UKRMP investigators to characterise other cell therapy approaches.

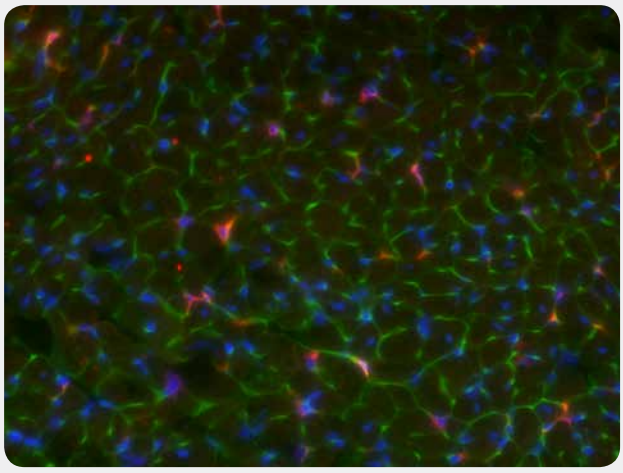
Supplementing the above research, our Hub investigators in Newcastle - James Shaw and his research associate Helen Marshall - are studying the effects of the immediate blood mediated inflammatory reaction (IBMIR) on complement component and Toll-like receptor (TLR) signalling following hepatocyte transplantation into the portal vein.

How does the inflammatory niche contribute to endogenous repair and influence the fate of transplanted cells?

To study endogenous repair, Fiona at KCL and Caetano Reis e Sousa at the Francis Crick Institute have provided transgenic mice to other investigators to study the roles of lineage-traced fibroblasts, macrophages and dendritic cells in modulating endogenous repair of various tissues. Using mice from Fiona Watt, research associate Matthias Friedrich from Fiona Powrie's laboratory at the University of Oxford is

studying the role of subpopulations of fibroblasts in colitis. **Sussane Sattler** a research associate in Sian Harding's laboratory has used the mice provided by Caetano to establish two cardiac inflammation models to study the role of a subpopulation of dendritic cells in endogenous cardiac regeneration and these models are currently being optimised for use both within the Hub and by other UKRMP researchers. In addition, personnel from the laboratories of Fiona Watt and Francesco Dazzi at King's, and Phil Newsome at Birmingham have been comparing the immune modulatory properties of mouse and human bone marrow derived PDGFRalpha +/Sca1+ mesenchymal stromal cells (MSCs) with PDGFRalpha +/Sca1+ skin fibroblasts isolated directly from adult dorsal skin.

Several known immune regulatory molecules have been revealed in the arrays and are currently being validated. Fiona Watt's group have been evaluating the PU.1YFP reporter mouse as a tool for detecting myeloid cells in the skin. She has found that labelled cells are readily detected by immunohistochemistry and flow cytometry. In skin the YFP+ cells are primarily macrophages and they increase in abundance during chronic inflammation and following skin wounding. Macrophages have been ablated using the CD11c-DTR model and this does not affect the rate of skin wound healing.



Mycordial tissue showing cell membranes labelled green, nuclei labelled blue and evenly scattered resident CD45+ immune cells labelled red.

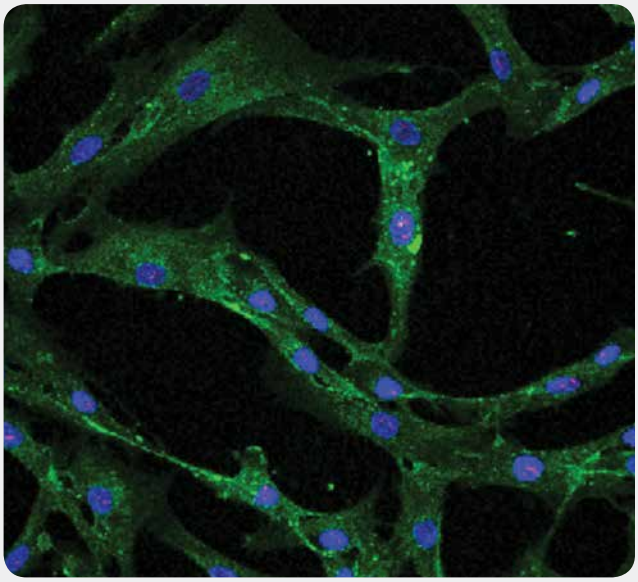
Hub Growth

The past year has seen expansion of Hub activities through two partnerships with the Acellular Technologies Hub in particular. Raul at King's is working with Derfogail Delcassian from the University of Nottingham (Acellular Hub), on a project which aims to create a replacement for regular, systemic drug delivery in the form of microparticles that can induce immune tolerance in transplant therapies.

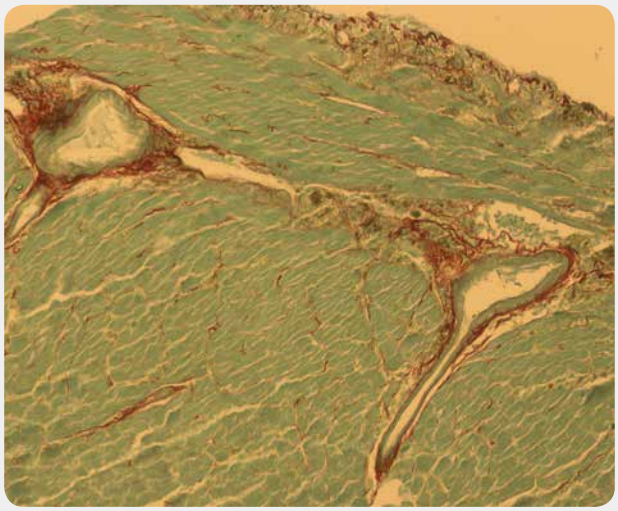
Additionally, a collaboration with Stefano Pluchino at University of Cambridge (and Acellular Hub) has been established to determine the immunomodulatory properties of extracellular vesicles (EVs). The collaboration is based on Stefano's studies on EV production by neural stem cells which showed that EVs possess immunomodulatory molecules. The current collaboration will investigate whether iPSC-derived hepatocytes, cardiomyocytes and RPE cells produce EVs containing the same immunomodulatory molecules and how these molecules might facilitate cell therapy approaches. As mentioned above, we have a new collaboration is with Marcus Dorner, comparing the behaviour of iPSC-derived and adult hepatocytes following transplantation into a humanised liver injury model.

Networking Activities

In December 2015, the Hub held a workshop titled Immunomodulation of Stem Cells at the Academy of Medical Sciences, London, which coincided with the official opening of the Centre for Stem Cells and Regenerative Medicine at KCL. Leaders representing industry and academia discussed the impact of the immune system in regenerative medicine and there was a well-attended poster session. Fiona Watt chaired a panel discussion (https://www.youtube.com/watch?v=3pQTJZM1O9c) involving Prof David Mooney (Harvard University), Dr Cathy Prescott (Biolatris Ltd), Dr Eleuterio Lombardo (TiGenix) and Prof Stuart Forbes (Niche Hub) on the commercialisation of cell therapies. The panel discussed ways of tackling challenges and creating opportunities in the field. A further panel discussion on the challenges of cell therapy trials in the UK in collaboration with RegMedNet is also available (https://www.youtube.com/watch?v=t56dUDlMjV0).



Mesenchymal stromal cells (green) from bone marrow. Nuclei are labelled blue.



Cardiac tissue from a mouse with immune-mediated heart damage showing fibrosis (red) surrounding vacuous blood vessels

Future Directions

In summary, the Hub is working towards a comprehensive understanding of how the immune system can be modulated to enhance cell therapies involving both cell transplantation and endogenous tissue repair.

Outputs

- Resources available to the community, see Section 4.

3. Disease Focused Projects

Second stage funding for the Platform is supporting five disease focused projects undertaking translational programmes in areas ripe for clinical development.

3.1 Professor Pete Coffey (University College London)
3.2 Dr David Hay (University of Edinburgh)
3.3 Dr Ilyas Khan (Swansea University)
3.4 Professor Andrew McCaskie (University of Cambridge)
3.5 Professor Manuel Salmeron-Sanchez (University of Glasgow)

3. Disease Focused Projects

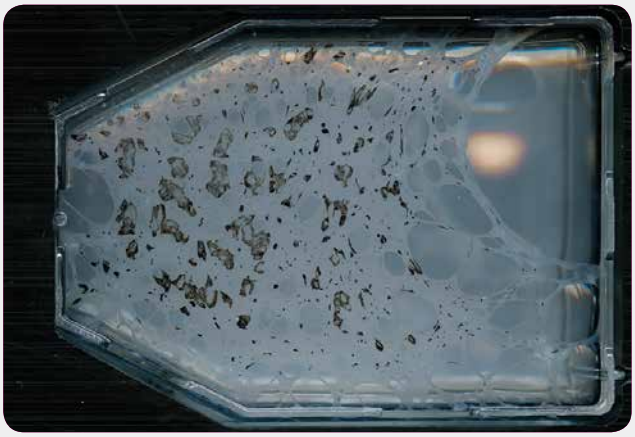
3.1 Professor Pete Coffey (University College London)

Scalable production of RPE cells from induced pluripotent stem cell under GMP conditions for cellular replacement therapy of the dry form of AMD.

The aim of this research is to expand our knowledge and generate sufficient preclinical safety data to support a Phase I/II clinical trial involving the cellular transplantation of human induced pluripotent stem cell (hiPSC)-derived retinal pigment epithelium (RPE) in patients with the dry form of age-related macular degeneration (AMD), a disease that is currently untreatable.

In the second year of this project, we have established the methods and protocols for producing clinical grade hiPSC from skin biopsies. Subsequently, we tested those hiPSC lines for unwanted integration of the vectors used for reprogramming, which could potentially raise safety issues. None were observed. The hiPSC lines were also sent to Cell behaviour, differentiation and manufacturing (PSCP) Hub of within the UKRMP where researchers at the Sanger Institute confirmed these findings using the most sensitive methods available. No adverse findings were detected in the three clinically manufactured lines that were tested.

Two patients were identified by Prof Lyndon da Cruz with dry AMD at Moorfields Eye Hospital who gave approval for skin biopsies to be taken in April. Those biopsies were transferred to the Royal Free Hospital GMP facility (Dr Mark Lowdell) for expansion and reprogramming (Dr Sajjida Jaffer). A further three patients will be identified and biopsies taken by the end of 2016. Following reprogramming and production of hiPSC lines, the patient lines will be differentiated into the therapeutic cells required to treat AMD – retinal pigmented epithelial cells (RPE).



RPE colonies forming from a flask of iPSCs

"Patients have consented to participate in the study and induced pluripotent stem cells are being produced from donated skin biopsies to manufacture therapeutic cells that can be transplanted back into those patients suffering from age-related macular degeneration."

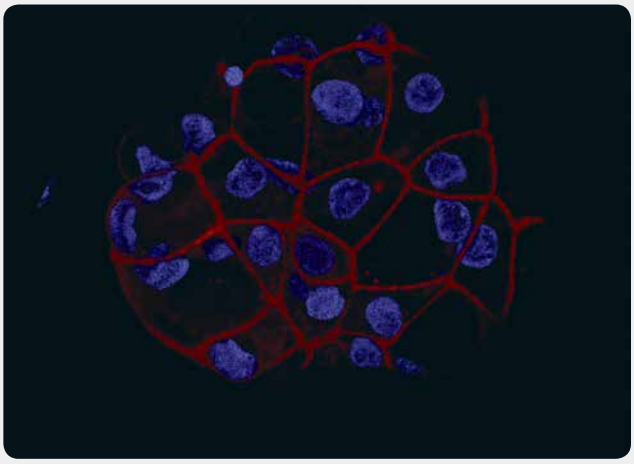
3.2 Dr David Hay (University of Edinburgh)

The development of 3-dimensional implantable liver organoids

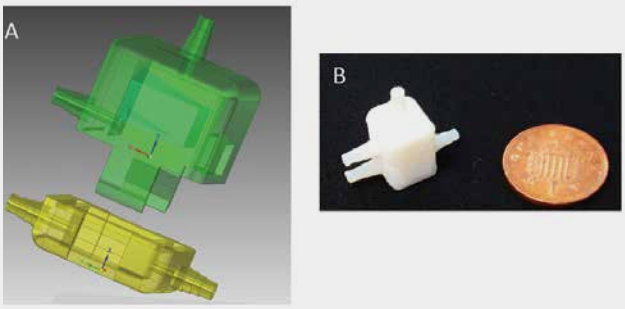
In the UK liver disease kills more people than diabetes and road deaths combined. The only curative option for end-stage disease is liver transplantation. However organ availability cannot meet demand and many patients die waiting for an organ, highlighting a clear need to develop scalable treatments. To address this, we have combined complementary scientific expertise and embarked on an interdisciplinary programme of translational research to build an artificial liver.

During the second year of the project, we have focused on optimising and characterising laboratory engineered liver tissue and a prototype mini-liver device. Importantly, laboratory generated tissue displays polarised features of human liver tissue. In contrast to the current gold standard of primary adult liver cells which are functionally stable for only 3-5 days in culture, stem cell engineered tissue displays stable function for over thirty five days, marking significant progress for the field.

More recently, we have studied the effect of blood flow on laboratory generated tissue. The results from these studies are encouraging with blood flow significantly improving liver cell phenotype towards that seen in human tissue, and leading to the reduction of immature liver and proliferative gene expression (Rashidi et al 2016, Archives of Toxicology). We have used the output from those studies and existing knowledge of human liver vasculature to refine artificial liver design, providing an appropriate niche for laboratory engineered tissue. In parallel to these projects, we have optimised our preclinical models of human liver disease and will begin testing the supportive nature of the laboratory generated tissue in the coming year. Exciting times ahead!



Liver tissue generated from a clinical grade human embryonic stem cell line, Man 12, expresses a key marker of cell polarity, zonal occuldin 1, in three dimensional culture.



The prototype human mini-liver have been designed with computer aided design and printed using a three dimensional printer.

"Prototype human liver tissue can be manufactured to specification and is stable in the laboratory for over thirty five days. This demon strates significant progress for the field and will be invaluable in artificial organ construction"

3.3 Dr Ilyas Khan (Swansea University)

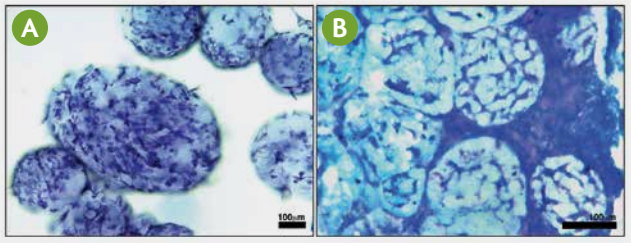
Generating durable and resilient repair of cartilage defects using tissue-specific adult stem cells – a systematic, therapeutic approach.

The major goal of this study is to produce functional osteochondral implants, and in keeping with the strategic aims of the Platform, to make their production amenable to scale-up. In the first year of this study we developed an accelerated and highly efficient method to differentiate articular cartilage-derived progenitor cells into cartilage, speeding up the process three-fold and making it more than 95% efficient. Our aim in the second year has been to develop strategies for rapid biofabrication of osteochondral constructs.

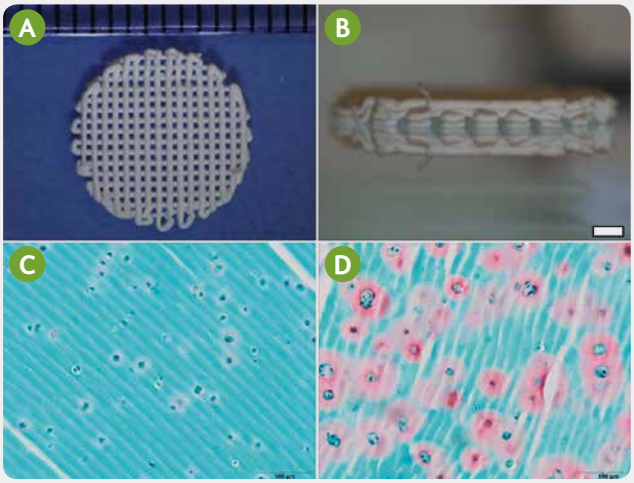
In order to efficiently expand chondroprogenitor cells to make cartilage we use porous microcarriers. Microcarriers are collagen-based similar in composition to the framework that provides the structural integrity of intact cartilage. Potentially microcarriers can act in a dual capacity, as surfaces for cell expansion and also as scaffolds for cartilage production. We have tested the latter proposition and showed that chondroprogenitors expanded on microcarriers can indeed form cartilage.

Working with colleagues at Utrecht Medical Centre (Professor Jos Malda) and Utrecht University (Professor Rene van Weeren) we are combining our expertise in chondrocyte biology and bioprinting to develop the methods for rapid biofabrication of osteochondral implants. Professor Malda's group are bioprinting calcium phosphate cement bases for implants upon which methycrylated collagen (GelMA) bioinks laden with chondroprogenitors are printed to successfully produce cartilage.

Our next major challenge is to produce implants that mimic the unique architecture of mature articular cartilage, so when they are implanted in patients they function as durable bearing surfaces.



Streamlining the biofabrication process. Chondroprogenitors grow efficiently on porous microcarriers (A). Cell-laden microcarriers can function as scaffolds for cartilage production, as shown in toluidine blue stained sections of microcarriers cultured in chondrogenic culture medium where extracellular matrix (purple/blue) can be found within and outside of microcarriers (B)



3D printed porous structures obtained with a calcium phosphate cement/hydrogel carrier for the bone component of the osteochondral graft (A-B). Safranin-O stained sections of bioprinted GelMA hydrogels with articular chondrocytes (C) and chondroprogenitors (D). Chondroprogenitors produce more extracellular matrix (red halos) than chondroctyes.

3.4 Professor Andrew McCaskie (University of Cambridge)

SMART STEP - Stepwise Translational Pathway for Smart Material Cell Therapy.

Osteoarthritis (OA) is a common disease that can ultimately destroy the surfaces of joints causing severe pain and reduced function. Current surgical treatments such as joint replacement are targeted to end stage disease, but surgical treatment options in earlier disease are limited. Our focus is on repair and regeneration of cartilage, which is the articular surface of a joint. The intervention would be at an early stage in order to reduce the progression of joint damage and hopefully delay the need for a joint replacement.

Within the adult human there are various cells that have the potential to bring about repair e.g. endogenous mesenchymal stem cells. Our approach is to target these cells and influence their behaviour using novel smart material technology together with the incorporation and controlled presentation of signalling molecules. Such a combination of a material and a molecule can change the behaviour of a cell by modulating signalling pathways to affect recruitment, proliferation and chondrogenic differentiation of endogenous mesenchymal stem cells – the key steps that might help repair cartilage. Our clinical goal is to make such treatments affordable, easy to apply and deliverable as a day case. Moreover, the Smart Step Pipeline establishes a stepwise translational pathway from "bench to bedside" to facilitate interaction with stakeholders beyond the consortium.

We have now completed our initial work to design and manufacture a scaffold material based on collagen in different specifications. We are now completing biological assessment of how cells react to the scaffold designs in order to select the best ones for clinical development This includes understanding how well cells cover the surface and travel into the structure. In terms of the molecules, we have generated, selected and validated a single cell-derived clone of Agrin expressing cells for consistent and optimal production. Protocols to separate the molecule from the cellular material have been successfulin vitro and are now being tested in vivo for toxicity. We will be taking selected combinations through pre-clinical development in the coming months.

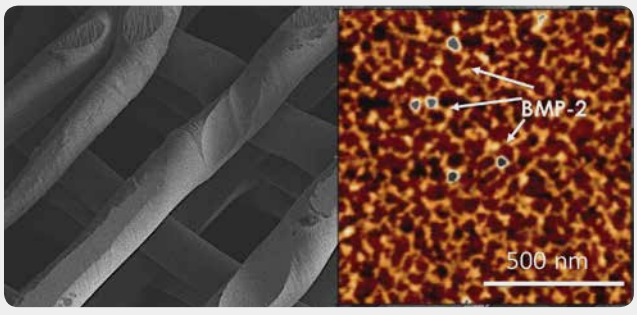
Scaffold and cell experiment shown actual size (A) and how it appears under a microscope view (B)

3.5 Professor Manuel Salmeron-Sanchez (University of Glasgow)

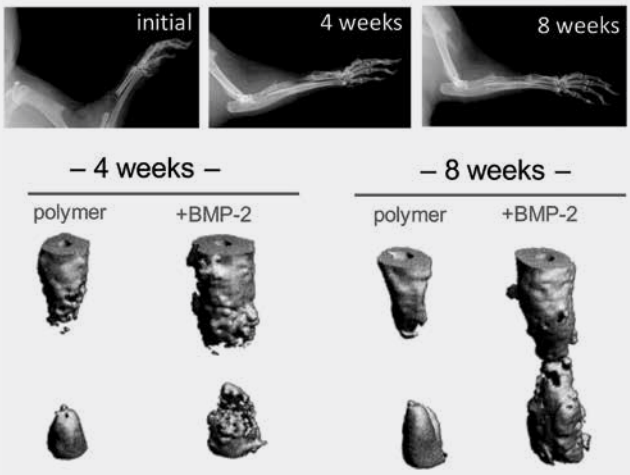
Synergistic microenvironments for non-union bone defects.

This project presents a therapeutic solution to address unmet clinical needs in bone regeneration in non-union bone defects. Our novel approach is based on the use of synthetic functional materials polymerised on the surface of 3D structural scaffolds. This simple, robust and translational material-based platform is being used for the safe and effective presentation of human growth factors (BMP-2 in particular) to engineer synergistic microenvironments to enhance bone regeneration.

During the second year of the project we have been focused on understanding our functional coatings on 3D printed scaffolds. These structural scaffolds are made of biodegradable polycaprolactone (3D printed fibres in a layer by layer cross deposition with pores ~ 500µm, in a collaboration with the University of Nottingham) and then coated with functional materials that bind growth factors very efficiently using plasma polymerisation and spray-drying technologies. We have fully characterised the bioactivity of BMP-2 on these surfaces and have optimised conditions to direct stem cell differentiation within the 3D systems. In addition, we have performed our first pre-clinical model in a critical size defect in the mouse radius, which has shown enhanced bone formation and full bridging of defects using very low concentrations of BMP-2 (~500 times lower than the current clinical standard 1.5 mg/ml).



Left, 3D printed biodegradable scaffolds coated functionalized to bind very low but effective doses of BMP-2. Right, the surface of the scaffolds with a fibronectin network and bound BMP-2.



Bone regeneration in a critical size defect in the mouse radius using materials functionalised to bind highly efficiently very low concentrations of BMP-2.

We have engineered 3D printed scaffolds coated with a functional polymer to present BMP-2 in a highly efficient way, allowing low growth factor concentrations, increased safety and reduced costs.

4. Hub Resources Available to the Community

4. Hub Resources Available to the Community

One of the aims of the UKRMP in overcoming the barriers to regenerative medicine being used in mainstream therapies is the development of new tools, reagents and protocols which can be utilised by the wider research community. By making such resources accessible to groups in both academic and industrial domains, it is anticipated that progress may be accelerated. Now in its third year of establishment, a growing number of such outputs are available through the Hubs.

These include the following:

Resource	Description	Hub	Contact	Further information
Tools and Reagents	Stem Cell Lines			
	MasterShef clinical grade human embryonic stem (hESC) lines	PSCP	Harry Moore h.d.moore@sheffield.ac.uk or via the UK Stem Cell Bank (UKSCB) enquiries@ukstemcellbank.org.uk	MasterShef (MShef) 01-09 hESC lines derived on Human Feeders in KOSR media. MShef 10 & 12 derived on Human Feeders in Nutristem, MShef11, 13 and 14 derived feeder-free in Nutristem media. Both clinical (EU-CTD compliant) and research grade versions are available - cell banks for the latter could also be made available through the UKSCB. http://www.nibsc.org/ukstemcellbank
	Mouse Lines			
	Pdgfr-α fibroblast-labelled mice	Immuno	Fiona Watt fiona.watt@kcl.ac.uk	Driskell RR et al. 2013. Nature, 504(7479):277-81
	Pu.1 macrophage-labelled mouse line	Immuno	Fiona Watt fiona.watt@kcl.ac.uk	Weber C et al. 2016. Cancer Res., 76(4):805-17.
	Clec9A+ dendritic cell-labelled mouse line	Immuno	Caetano Reis e Sousa Caetano@crick.ac.uk	Schrami B et al. 2013. Cell, 154(4):843-58.
	NOD/SCIDγc-/- humanised mouse line	Immuno	Giovanna Lombardi giovanna.lombardi@kcl.ac.uk	Xiao F et al. 2016. Br J Pharmacol., 173(3):575-87
	Humanised Fah-/- mouse line	Immuno	Marcus Dorner m.dorner@imperial.ac.uk	Billerbeck E et al. 2016. J Hepatol. 65(2):334-43.

Resource	Description	Hub	Contact	Further information
Tools and Reagents	Disease Models			
	Ovine medial femoral condyle defect model for bone repair	Acellular	Jane McLaren Jane.mclaren@nottingham.ac.uk	McLaren et al. Eur Cell Mater. 2014 Jun 8;27:332-49
	Murine and ovine models for bone formation	Acellular	Janos Kanczler J.Kamczler@soton.ac.uk	Tayton E et al. J Biomed Mater Res A. 2015 Apr;103(4):1346-56. doi: 10.1002/jbm.a.35279. Epub 2014 Jul 23.
	Ex vivo bone formation and angiogenic models in chick	Acellular	Janos Kanczler J.Kamczler@soton.ac.uk or Robin Rumney R.M.Rumney@soton.ac.uk	Smith EL et al. Eur Cell Mater. 2013 Sep 11;26:91-106; discussion 106. Review
	Optimised Isoperotonol and resiquimod mouse models of cardiac inflammation	Immuno	Susanne Sattler s.sattler@imperial.ac.uk	Information available upon request
	Cell labelling and delivery reagents			
	Super Paramagnetic Iron Oxide Nanoparticles (SPIONS) for labelling and tracking macrophages and stem cells	Safety	Mike Barrow mikesyb@liverpool.ac.uk	Barrow et al. Contrast Media Mol Imaging. 2016 Jun 30. doi: 10.1002/cmmi.1700
	Silica coated Gold Nanorods (GNRs) for labelling and tracking macrophages and stem cells	Safety	Raphael Levy rapha@liverpool.ac.uk	Comenge J et al. ACS Nano. 2016 Jun 20. doi: 10.1021/acsna no.6b03246
	Lentivirus plasmids • 2nd generation lentivirus vector pHIV-FP720-E2A-luciferase. • pHIV-Tyrosinase-eGFP (as a fusion protein) • pHIV-Tyrosinase-eGFP-IRES-Luciferase • pHIV-Tyrosnase-IRES-Luciferase • MBP-iR-FP720-E2A-Luciferase vector	Safety	Toni Plagge plagge@liverpool.ac.uk	For bicistronic expression of iRFP720 fluorescent protein and firefly luciferase via an E2A element from the EF1 alpha promoter (also available with an IRES element replacing E2A) Functionally tested in HEK293 cells Functionally tested in HEK293 cells Functionally tested in HEK293 cells Vector has the Myelin-basic-protein promoter instead of the generally active EF1a promoter. This promoter drives expression of the reporter genes in oligodendrocytes (cells that express the myelin basic protein).

Resource	Description	Hub	Contact	Further information
Tools and Reagents	Validated extracellular matrix (ECM) arrays	Niche	Sue Kimber sue.kimber@manchester.ac.uk	The Hub is producing a range of high-quality ECM products in order to support the development of bespoke niche products for preclinical research and therapeutic development.
	Biofunctionalised cryptic extracellular matrix to target epithelial to mesenchymal transition	Acellular	Benjamin Pierce b.pierce@imperial.ac.uk	Horejs c et al. Proc Natl Acad Sci U S A. 2014 Apr 22;111(16):5908-13. doi: 10.1073/ pnas. 1403139111. Epub 2014 Apr 3.
	Porous collagen scaf folds and modifiable hydrogels for articular cartilage repair.	Acellular	Benjamin Pierce b.pierce@imperial.ac.uk	Parmar PA et al. Biomaterials. 2015 Jun;54:213-25. doi: 10.1016/j.bio materials. 2015.02.079. Epub 2015 Apr 11. Parmar PA et al. Adv Healthc Mater. 2016 Jul;5(13): 1656-66. doi: 10.1002/ adhm.201600136. Epub 2016 May 24. Parmar PA et al. Biomaterials. 2016 Aug; 99:56-71. doi: 10.1016/ j.biomaterials.2016.05.011. Epub 2016 May 10.
	Porous PLGA microspheres for use as injectable cell carriers	Acellular	Omar Qutachi omar.qutachi@nottingham.ac.uk	Qutachi et al. Acta Biomater. 2014 Dec;10(12):5090-8. doi: 10.1016/j.actbio.2014.08.015. Epub 2014 Aug 23
Protocols	Cost effective protocols for growing hepatocyte-like cells from human pluripotent stem cells suitable for mass production of clinical grade cells	Niche	Dave Hay dave.hay@talktalk.net	Stem Cell Reports. 2015; 5 (5): 1250-1262. doi: 10.1016/j.stem cr.2015.10.016. PMID: 26626180
	Techniques for measurement of lead microRNAs in patients with acute liver injury	Niche	James Dear james.dear@ed.ac.uk	Nature Scientific Reports 5, Article number: 15501 (2015) doi:10.1038/srep15501
	Raman spectroscopy protocol for integration and analysis of multiple analytical datasets	Niche	Ben Pierce b.pierce@imperial.co.uk	J. Biophotonics 2016; 9 (5), 542–550doi 10.1002/ jbio.201500238

Resource	Description	Hub	Contact	Further information
Technologies	Acellular			
	Fabrication and subsequent culture of tubular tissues	Acellular	James Dixon james.dixon@nottingham.ac.uk	Othman et al. Biofabrication. 2015 Apr 14;7(2):025003. doi: 10.1088/1758-5090/7/2/025003
	3D printed scaffolds and 3D bioprinting of constructs for bone repair	Acellular	Jing Yang jing.yang@nottingham.ac.uk Felicity Rose Felicity.rose@nottingham.ac.uk	Ruiz-Cantu L et al. Biofabrication. 2016 Mar 1;8(1):015016. doi: 10.1088/1758-5090/8/1/015016. Sawkins MJ et al. Biofabrication. 2015 Jul 2;7(3):035004. doi: 10.1088/1758-5090/7/3/035004.
	Safety			
	Methods for detecting common genetic changes in PSC cultures	PSCP	Ivana Barbaric i.barbaric@sheffield.ac.uk	Human pluripotent stem cells (hPSCs) can adapt to in vitro conditions by acquiring non-random genetic changes that render them more robust and easier to culture (eg trisomies of chromosomes 1, 12, 17 and 20). hPSCs should therefore be regularly screened for such aberrations but this necessitates a good understanding of the sensitivities of different methods used. An assessment has been made to understand the limits of mosaicism detection by commonly employed methods such as chromosome banding, quantitative PCR, fluorescent in situ hybridization and digital droplet PCR. Baker D et al. Stem Cell Reports (submitted 29th March 2016 - under consideration).
	Random sequence control material for detection of viral contamination via Next Generation Sequencing (NGS).	PSCP	Glyn Stacey Glyn.Stacey@nibsc.org	An evaluated set of potential control materials and procedures for use in optimisation and control of NGS detection of adventitious agents.

Resource	Description	Hub	Contact	Further information
Technologies	Screening			
	Tools for drug toxicity screening based on stem cell derived hepatocytes Screening strategies for remyelination	Niche	Dave Hay dave.hay@talktalk.net	Stem cell derived liver tissue for transplant and human safety screening Cameron et al. Stem Cell Reports. 2015 Dec 8;5(6):1250-62. doi: 10.1016/j.stemcr.2015
	Screening strategies for remyelination	Niche	Anna Williams anna.williams@ed.ac.uk	Exp Neurol. 2011 Jul;230(1):138-48. doi:10.1016/ j.expneurol.2011.04.009. PMID:21515259
Data sets	Whole Genome Sequencing, RNASeq and Bisulphate Sequencing of 2 hESC lines; and sub-clonal hESC derivative lines	PSCP	Peter Andrews p.w.a@sheffield.ac.uk	A total of 80 sub-clonal lines from single clones of both MShef4 and MSheff11 hESC lines have been squenced (Whole Genome, Bisulphate and RNAseq) to assess mutation rates. These clones and their sequence are available to qualified investigators for further study.
	Immunoprofiles of: • iPSC-derived hepatocytes • retinal pigment epithelial (RPE) cells • cardiomyocytes	Immuno	Raul Elgueta (hepatocytes) raul.elgueta@kcl.ac.uk Giorgia Fanelli (RPE) giorgia.fanelli@kcl.ac.uk Fang Xiao (cardiomyocytes) fang.xiao@kcl.ac.uk	Information available upon request
Equipment	Microscopy			
	Microscope Slide Scanner Media Cybernetics	Niche/ CCBN*	Alex Raven s1351928@sms.ed.ac.uk	http://www.crm.ed.ac.uk/ equipment/micro scope-slide-scanner
	Raman Microscope Renishaw InVia	Niche	Ben Pierce b.pierce@imperial.co.uk	http://www.imperial.ac.uk/ vibrational-spectroscopy-and-chemical-imaging/ facilities/raman-spectrometers/
	Raman Microscope Renishaw InVia	Niche/ CCBN	Colin Campbell colin.campbell@ed.ac.uk	http://www.crm.ed.ac.uk/ equipment/renishaw-invia-raman-microscope
	Photothermal microscope, and cell tracking velocimeter Fluorescent lightsheet microscope.	Safety	Raphael Levy rapha@liverpool.ac.uk	https://www.liverpool.ac.uk/ integrative-biology/ facilities-and-services/ centre-for-cell-imaging/

* The Computational and Chemical Biology of the Stem Cell Niche

Resource	Description	Hub	Contact	Further information
Equipment	Imaging			
	Operetta High content imaging	Niche/ CCBN	Eoghan O'Duibhir eoghan.oduibhir@ed.ac.uk	http://www.crm.ed.ac.uk/equipment/operetta-high-content-microscope
	Non-destructive cell imaging platform applicable in bone and cartilage regeneration research	Niche	Pierre Bagnaninchi Pierre.Bagnaninchi@ed.ac.uk	
	9.4T MRI, benchtop 1T MRI, SPECT/CT, PET/CT, photoacoustic, ultrasound, biolumi nescence, X-ray CT	Safety	Tammy Kalber t.kalber@ucl.ac.uk	http://www.ucl.ac.uk/cabi
	9.4T MRI scanner, MSOT photoacoustic, IVIS bioluminescence, ultrasound	Safety	Harish Poptani harishp@liverpool.ac.uk	https://www.liverpool.ac.uk/translational-medicine/research/centre-for-preclinical-imaging/
	SQUID magnetometer,	Safety	Claire Hutchinson claire.hutchinson@liverpool.ac.uk	Barrow et al. Biomater. Sci., 2015,3, 608-616 doi:10.1039/C5BM00011D
	7T MRI, 3T benchtop MRI, bioluminescence, PET	Safety	Steve Williams steve.williams@manchester.ac.uk	http://research.bmh.manchester.ac.uk/imaging
	Phase imaging microCT, serial block face SEM imaging and light sheet microscopy	Acellular	Richard Oreffo Richard.oreffo@soton.ac.uk Anton Page A.Page@soton.ac.uk	Southampton Imaging http://www.southampton.ac.uk/microscopy/index.page Xradia XRM-410 Phase enhanced high resolution µCT Gatan 3-view microscope and LaVision Ultramicroscope light sheet microscope.

Resource	Description	Hub	Contact	Further information
Equipment	Manufacture			
	DB FACS Aria III Fusion, High speed cell sorter	Niche	Fiona Rossi fiona.rossi@ed.ac.uk	http://www.crm.ed.ac.uk/equipment/bd-facs-aria-iii-fusion
	Electrospinner IME Technologies	Niche/ CCBN	Siobhán Dunphy s.dunphy@ed.ac.uk	http://www.crm.ed.ac.uk/equipment/ime-electrospinning-device
	Femtosecond Laser 3D structure fabrication	Niche/ CCBN	Robert Thomson r.r.thomson@hw.ac.uk	http://www.crm.ed.ac.uk/equipment/femtosecond-laser
Workshop Reports/ Papers	2015 Assessment of Source Materials for Cell Based Medicines Workshop Report	PSCP/ Safety	Glyn Stacey Glyn.Stacey@nibsc.org	Submitted to Stem Cell Translation Medicine
	2015 Comparability Workshop Report	PSCP	David Williams D.J.Williams2@lboro.ac.uk	Regen Med. 2016 Jul;11(5):483-92. doi: 10.2217/rme-2016-0053. Epub 2016 Jul 12
	2015 Nanoparticles Workshop Report	Safety	Raphael Levy rapha@liverpool.ac.uk	In preparation
Services	Stem cell cytogenetics - diagnostics and characterisation	PSCP	Duncan Baker duncan.baker@sch.nhs.uk	https://www.sheffieldchildrens.nhs.uk/our-services/sheffield-diagnostic-genetics-service/laboratory-services.htm#cytogenetic

5. Hub/Industry Interactions and Case Studies

5. Hub/Industry Interactions and Case Studies

Engagement with industry is a key objective of the UKRMP. Linkage will in part be achieved through the utilisation of the tools and technologies generated by the Hubs, but longer term relationships are also being built across the Platform. The establishment of these new partnerships will allow the Hubs to develop innovative technological solutions aligned with end-user requirements and in so doing will provide the impetus for commercialisation and ultimately the clinical application of regenerative medicine technologies.

A brief overview of Hub activities or individual case studies are presented below:

PSCP Hub

PSCP's engagement with industry has primarily focused on providing opportunities to raise, discuss and try to resolve bottlenecks within the translation of regenerative medicine. At the "Comparability Workshop" held in Cambridge in September 2015, twenty-nine developer delegates attended from more than twenty different companies from three continents, including large biopharma representatives from GSK and Neusentis (Pfizer ltd), SMEs such as Reneuron and Autolus Limited, reagent suppliers including Ajinomoto, and equipment providers including TAP Biosystems, Fujifilm and FloDesign Sonics. Alongside these intermediate institutes such as IStem and IBMT Fraunhofer, and standards organisations NIBSC, LGC Group and NIST, were also represented. This workshop, and preparation of the white paper outcome, also involved significant and supportive interaction with the MHRA.

PSCP researchers are continuing to provide services, both nationally and internationally, to developers of therapeutics and machine suppliers relating to process and facility design, particularly for the scale out of the production of therapeutics especially using automated expansion and differentiation platforms. Furthermore, through its partnership projects, PSCP is addressing issues of cell–therapy contract manufacture and cross-site comparability using commercially available equipment and assessing process variation. This work uses the TAP Biosystems automation platform.

The PSC-derived dopaminergic neuron research programme is working extensively with the Cell and Gene Therapy Catapult to develop a business case for taking forward the hPSC-derived dopaminergic neuron therapy. The CGT Catapult is assisting with health economic issues, patent and IP work and will be assisting with the compilation of the clinical trials dossier. Further funding is being sought to continue this collaboration in partnership with two PSCP collaborators for continued product development and pre-clinical testing.

PSCP is also involved in collaborative working with supplier companies such as Ajinomoto, Biolamina and Miltenyi to test and develop reagents and assays which will assist with regulatory compliance and therefore, the translation of PSC-derived therapies from bench to clinic.

Finally, PSCP is making a number of contributions to the development of international standards, particularly with the activities of the ISO TC 276 group on Manufacturability.

Niche Hub – Case Study - Dr David Hay, University of Edinburgh

The UKRMP platform has provided the Hay laboratory with support to develop renewable sources of human liver tissue for translational medicine. The project is interdisciplinary in nature and links a number of the UK's strengths in regenerative medicine. The aim of this research is to use the in vitro derived tissue for cell based modelling 'in a dish' and to develop pioneering cell based therapies for human liver disease. Interest lies specifically in the major cell type of the human liver, the hepatocyte. Hepatocytes perform multiple functions within the liver making this cell type an important target to study.

During the project the team has developed defined and semi-automated systems for differentiating hepatocyte-like cells from pluripotent stem cells. The prototype systems have been tested on research and clinical grade human embryonic stem cells in collaboration with the University of Manchester. Those systems have demonstrated equivalence to human hepatocytes when we examine protein secretion, prescription drug metabolism and predict compound potential to cause drug induced liver injury. Given the importance of these results the Hay lab has protected the GMP ready method for cell production in partnership with the extracellular matrix provider, Biolamina. More recently the team has partnered with AstraZeneca to screen compound libraries to examine their effect on hepatocyte and liver biology. As this work with cell based screening moves forward it is anticipated that this will provide a better understanding of the potential of in vitro derived tissue and how it can be improved and appropriately deployed in vivo to treat human liver disease. In that vein, the Hay group is collaborating with King's College London to optimise the encapsulation of stem cell derived hepatocytes using a GMP compatible process that have a successful track record in the clinic.

Safety Hub

The Safety and Efficacy Hub has been collaborating with iThera Medical since 2013. The company offers the only opto acoustic imaging system (MSOT - multispectral optoacoustic tomography) with real-time whole-body small animal imaging capability. The instrument can effectively image cells labelled with gold nanorods, or that have been modified to express a near infrared fluorescent protein (iRFP720). By labelling iRFP-expressing cells with gold nanorods, it is possible to locate the administered cells over the short and long-term with a great degree of precision, thus providing accurate biodistribution data. This is important for assessing the safety of the cells, as it can be determined if the cells engraft in non-target organs and/or form tumours in the longer term. In addition, by monitoring the clearance kinetics of indocyanine green (ICG) and IRDye800 caroboxylate, it is possible to monitor liver and kidney function, respectively, in individual animals over a time course. This ability to monitor organ function longitudinally will enable us to accurately assess the efficacy of RMTs.

The MSOT system at the Safety Hub is now the most utilised worldwide and through data analysis, modelling, and image reconstruction. Outputs from this fruitful collaboration include three publications in the past 12 months, Scarfe et al, Comenge et al, Sharkey et al, (see Annex 4) plus the establishment, in 2014, of a UK MSOT user group with iThera, Leeds and Cambridge Universities to build an effective UK community which enables sharing of best practices and protocols. iThera have continued collaboration with Hub members resulting in both a successful local Knowledge Exchange award, where they provided support in data analysis in liver disease models, and also partnered a small EPSRC-funded grant focussed on developing multifunctional SPION-NIR particles. Furthermore, iThera have contributed to workshops and symposiums with continued interaction and discussion.

Acellular Hub – Case Study - Dr James Dixon, University of Nottingham

One of the main stumbling blocks in regenerative medicine is the inefficient delivery of targeted therapies. With the emergence of gene editing and reprogramming techniques, enhanced ways of targeting cells are needed which will allow us to directly correct genetic mutations, such as those seen in muscular dystrophy, cystic fibrosis and cancer.

Within the Acellular technologies Hub we have developed a highly efficient system using small proteins that target the cell's sugar coating (heparan sulphate). The tethered therapeutic molecules are delivered at high levels inside cells meaning they have much more effect at significantly lower doses. This approach is called GET (Glycosaminoglycan-enhanced transduction) technology.

GET technology has been patented and licenced by Locate Therapeutics (a University of Nottingham Spin-out SME) for application to orthopaedic regenerative medicine. Here the technology will be integrated with other patented technology to enhance bone healing using biodegradable implants. The team is actively pursuing many other regenerative medicine applications including GET to reprogramme differentiated cells to stem cells (iPSCs), programme cells to clinically relevant cell-types for therapy (eg. cardiomyocytes) and to enhance gene-editing platforms (CRISPR/Cas9). Within the UKRMP initiative collaborations have been established that aim to use GET in tracing stem cells post-implantation, both non-invasively and without genetic modification (with the Safety hub). GET is also being used to create gene-activated scaffolds for skeletal tissue engineering (with Professor Fergal O'Brien, RCSI, Ireland) and due to the nature of GET (targeting heparan suphate) also shows promise for developing cell-type specific delivery methods for cancer targeting.

To move rapidly towards clinical translation the Dixon team is collaborating with John's Hopkins University, USA (Prof. Justin Hanes and Jung Soo Suk) to develop in vivo delivery of therapies, specifically to the brain and lung. Proof-of-principle has been obtained to show that GET can efficiently mediate gene-therapy in the mouse lung and this will now be extended to create a genetic medicine which could tackle a number of lung disorders, including cystic fibrosis (in collaboration with members of the UK Cystic Fibrosis gene therapy consortium). Through this collaboration GET delivery will hopefully become one of the first UKRMP core technologies to reach first-in-man trials.

Immunomodulation Hub

The Hub continues to forge links with industrial companies with the aim of future collaboration. At the end of November 2016 the Hub will be hosting a workshop at King's College London on the clinical applications and opportunities arising from mesenchymal stroll cell (MSC) research. This workshop will feature strong participation by industry–most notably TiGenix and Biolatris, who were instrumental in shaping the panel discussion that took place at the at the Immunomodultion of Stem Cells meeting in London last year. This workshop will not only provide an opportunity to learn about opportunities and applications within the MSC research field, but also an opportunity to seek advice regarding the development of the Hub's own clinically translational outputs and successful business models for commercialisation. Giovanna Lombardi has a long-standing relationship with Miltenyi Biotech, which focuses on optimising GMP-compliant protocols for Treg isolation and expansion for forthcoming clinical trials. The Hub is currently in discussion with Miltenyi Biotech on ways to utilise this expertise for ongoing work in Fiona Watt's and Francesco Dazzi's laboratories that are investigating using fibroblast subpopulations and MSCs respectively for cell therapy approaches. Miltenyi will also be a key participant at the MSC workshop.

6. UKRMP Special Merit Prize

6. UKRMP Special Merit Prize

This past year two UKRMP special merit prizes have been awarded to acknowledge and reward Hub post-doctoral researchers who have demonstrated outstanding activity in providing connectivity across the Hubs and Platform to deliver its mission.

The prizes were awarded to:

- Dr Mads Bergholt, Niche Hub (Imperial College London)
- Dr Michael Barrow, Safety Hub (University of Liverpool)

In both cases, Mads and Mike were awarded the prize based upon their proactive approach to their research which fully aligned with the Platform's philosophy of promoting interdisciplinary team science. In doing so they both identified opportunities and demonstrated creativity by adopting novel approaches in leading the implementation of their activities to achieve maximum impact. In short, they went beyond 'business as usual' and embraced the collaborative Hub ethos to enhance delivery of the objectives of the Hubs and Platform.

Dr Mads Bergholt

Dr Michael Barrow

Mads' activities centred around the application of Raman spectroscopy – a technique used to understand more about the composition of material–to investigate stem cell behaviour and within regenerative medicine. Mike's activities were built around his synthesis of novel supraparamegnetic iron oxide nanoparticles (SPIONs) for cell tracking using MRI and their distribution for use across the Platform.

This is an annual competition, which will run again next year, with nominations made by the Hub Directors.

Annexes

Annex 1
UKRMP governance

Annex 2
UKRMP Hub awards
UKRMP disease focused projects
MRC regenerative medicine capital awards

Annex 3
UKRMP Hub research teams

Annex 4
Hub publications list

Annex 1

UKRMP Governance

Executive Group

- **Dr Rob Buckle,** Director of Science Programmes, MRC; Director UKRMP
- **Professor Ian Greer**, Vice-President and Dean, Faculty of Medical and Human Sciences, The University of Manchester; Chair UKRMP Programme Board
- **Dr Declan Mulkeen,** Chief Science Officer, MRC
- **Dr Annette Bramley,** Head, Healthcare Technologies, EPSRC
- **Dr David Pan,** Programme Manager UKRMP
- **Professor Melanie Welham,** Chief Executive, BBSRC

Programme Board

- **Professor Ian Greer** (Chair), University of Manchester, UK
- **Professor Frances Balkwill,** Queen Mary University of London, UK
- **Professor Nissim Benvenisty,** The Hebrew University of Jerusalem, Israel
- **Professor Kenneth Boheler,** University of Hong Kong, China
- **Dr Drew Burdon,** Smith and Nephew, UK
- **Dr Nigel Burns,** Cell Medica, UK
- **Professor Jöns Hilborn,** Uppsala University, Sweden
- **Dr Trevor Howe,** Janssen R&D, Belgium
- **Dr Andrew Lynn,** University of Cambridge, UK
- **Professor Marc Peschanski,** I-STEM Paris, France
- **Professor Dr Petra Reinke,** Berlin-Brandenburg Centre for Regenerative Therapies, Germany
- **Professor Anne Rosser,** Cardiff University, UK
- **Professor Paul Whiting,** Alzheimer's Research UK, UCL Drug Discovery Institute, UK
- **Professor Peter Zandstra,** University of Toronto, Canada

Annex 2

UKRMP Hub awards

- Professor Peter Andrews, University of Sheffield
  Cell behaviour, differentiation and manufacturing Hub (£4.6M)
  Partnership programmes included within main award:
  - *Development of GMP ES cell derived dopaminergic neurons in preparation for a first in human clinical trial in Parkinson's Disease*
  - *Comparability of automated expansion of PSC at three international sites*
  - *The consequences of cryptic genetic variants in cultures of human Pluripotent Stem Cells for safety and efficacy of applications for regenerative medicine – PSCP/Safety Hubs and Stage II Coffey Project*

- Professor Stuart Forbes, MRC Centre for Regenerative Medicine, University of Edinburgh
  Engineering and exploiting the stem cell niche Hub (£4.6M)
  Partnership programmes included within main award:
  - *ECM matrix products for niche biomaterials and biology*
  - *New liver microRNA toxicity biomarkers – Niche/Safety Hubs*
  - *Delivering a niche for liver repair and chondrocyte differentiation – Niche/Acellular Hubs*
  - *ECM and Wnt interactions of human iPSC-derived hepatocytes*
  - *Defining a translational niche for tissue engineered products*

- Professor Kevin Park, MRC Centre for Drug Safety Science, University of Liverpool
  Safety and efficacy, focussing on imaging technologies Hub (£4.6M)
  Partnership programmes included within main award:
  - *Evaluation of the safety and efficacy in a novel preclinical therapy - regeneration of damaged renal tissue within donor kidneys*
  - *Development of novel cell tracking probes for nuclear and optical/photoacoustic imaging*
  - *Mechanistic biomarkers that guide the safe and effective utilisation of regenerative medicine therapeutics for liver fibrosis*
  - *Magnetic targeting of therapeutic cells for enhanced efficacy and safety of liver fibrosis treatment*
  - *Assessment of the tumorigenic potential of a frequent ES cell genetic variant, 20q.11.21 amplicon, in a liver engraftment model; Safety/PSCP/and Niche Hubs*
  - *Evaluating the biodistribution and toxicity of a pluripotent stem cell-based therapy for Parkinson's disease – Safety/PSCP Hubs*

- Professor Kevin Shakesheff, University of Nottingham
  Acellular approaches for therapeutic delivery Hub (£3.8M)
  Partnership programmes included within main award:
  - *New materials:*
    - *i. Extracellular vesicles (EV) that deliver mRNA*
    - *ii. Self-assembling peptides that responsively change local elasticity*
  - *New materials for clinical applications:*
    - *i. Microparticles for cell and drug delivery*
    - *ii. Liposomal systems for dentine regeneration*
    - *iii. A thin, rollable and transparent gel matrix for corneal endothelial cell transplantation*
    - *iv. Development of fibrous material for cell delivery in the eye and tendon*
  - *Drug delivery systems to enhance engraftment of cells – Acellular/Niche Hubs*
  - *Biomaterial-based approaches to deliver extracellular vesicles for cardiac tissue repair*
  - *Development of a medical device to support the delivery of cell therapies in surgery*

- Professor Fiona Watt, King's College London
  Immunomodulation Hub (£2.3M)
  Partnership programmes included within main award:
  - *Micro-particles for the induction of immune modulation in the transplant niche – I mmunomodulation/Acellular Hubs*
  - *Dissecting the molecular function of stem cell-derived extracellularvesicles (EVs) in educating the host inflammatory niche – Immunomodulation/Acellular Hubs*

UKRMP disease focused awards

- Dr Ilyas Khan/Professor Charles Archer, Swansea University.
  Generating durable and resilient repair of cartilage defects using tissue-specific adult stem cells – a systematic, therapeutic approach. £1M * (£0.29M RC, £0.2M ARUK, Reumafonds £0.51M)
- Professor Pete Coffey, University College London
  Scalable production of RPE cells from induced pluripotent stem cell under GMP conditions for cellular replacement therapy of the dry form of Age-related macular degeneration (AMD). £1.6M
- Dr David Hay, MRC Centre for Regenerative Medicine, University of Edinburgh
  The development of 3 dimensional implantable liver organoids. £1.6M
- Professor Andrew McCaskie, University of Cambridge
  (SMART STEP) Stepwise Translational Pathway for Smart Material Cell Therapy. £1,6M * (£0.64M RC, £0.53M ARUK, Reumafonds £0.43M)
- Professor Manuel Salmeron-Sanchez, University of Glasgow
  Synergistic microenvironments for non-union bone defects. £1M # (£0.54M RC, £0.46M ARUK)

* *partnered with Arthritis Research UK and Reumafonds*
\# *partnered with Arthritis Research UK*

MRC regenerative medicine capital awards

UKRMP-linked

- Professor Peter Andrews, University of Sheffield. Pluripotent Stem Cell Platform - Capital Investment, £3.1M
- Professor Cay Kielty, University of Manchester. Regenerative medicine: instrumentation for flow cytometry and cell printing. £0.7M
- Professor Stuart Forbes, University of Edinburgh. The Computational and Chemical Biology of the Stem Cell Niche, £5.0M
- Professor Sheila MacNeil, University of Sheffield. Open-access biomaterials microfabrication and non-invasive imaging facilities for Regenerative Medicine, £0.7M
- Professor Richard Oreffo, University of Southampton. Southampton Imaging: 3D imaging at millimetre to nanometre scales for regenerative medicine using multiple complimentary modalities, £1.2M
- Professor Brian Park, University of Liverpool. In vivo imaging technologies to assess the efficacy and safety of regenerative medicine therapies, £3.3M
- Professor Molly Stevens, Imperial College London. State of the Art Biomaterials Development and Characterization of the Cell-Biomaterial Interface, £1.2M

Capital awards out with the UKRMP Hubs

- Professor Raimondo Ascione, University of Bristol. Pre-clinical In-vivo Functional Imaging for Translational Regenerative Medicine, £2.8M
- Professor Robin Ali, University College London. A flow cytometry facility for ocular regenerative medicine, £0.7M
- Professor Anne Dickinson, Newcastle University. Clinical grade cell separation technologies in the Newcastle Cellular Therapies Facility, £0.2M
- Professor Sian Harding, Imperial College London. BHF Imperial Cardiovascular Regenerative Medicine Centre, £0.7M
- Dr Charles Hunt, UK Stem Cell Bank (NIBSC). Automation of Cell Banking & Characterisation Pathways at the UKSCB: Underpinning Delivery of a Core Component of the UK Infrastructure for Regen Med, £0.3M

Annex 3

UKRMP Hub research teams

(outside of Principal and Co-Investigators; listed in respective Hub sections)

PSCP Hub

- **Dr Elsa Abranches**, National Institute for Biological Standards and Controls
- **Mr Duncan Baker**, University of Sheffield
- **Dr Nick Blair**, University of Cambridge
- **Dr Amit Chandra**, Loughborough University
- **Ms Mercy Danga**, University of Cambridge
- **Miss Sian Gregory**, University of Sheffield
- **Dr Ross Hawkins**, National Institute for Biological Standards and Controls
- **Dr Marta Milo**, University of Sheffield
- **Dr Serena Nik-Zainal**, The Wellcome Trust Sanger Institute, Cambridge
- **Dr Venkat Pisupati**, University of Cambridge
- **Mr Allan Shaw**, University of Sheffield
- **Dr Sujith Sebastian**, Loughborough University
- **Dr Oliver Thompson**, University of Sheffield
- **Dr Loriana Vitillo**, University of Cambridge
- **Dr Ferdinand von Meyenn**, The Babraham Institute, Cambridge
- **Mr Andrew Wood**, University of Sheffield

Niche Hub

- **Dr Wei-Yu Lu**, University of Edinburgh
- **Dr Kate Cameron**, University of Edinburgh
- **Dr Eva Borger**, University of Edinburgh
- **Dr Holger Schulze**, University of Edinburgh
- **Eleojo Obaje**, University of Edinburgh
- **Dr Wesam Gamal (Sam)**, University of Edinburgh
- **Dr Chao Li**, University of Liverpool
- **Dr Mads Bergholt**, Imperial College London
- **Dr Jean-Phillipe St-Pierre**, Imperial College London
- **Dr Andrea Serio**, Imperial College London
- **Dr Mike Albro**, Imperial College London
- **Dr Yvonne Reinwald**, Keele University
- **Sebastiaan Zijl**, King's College London
- **Dr Stuart Cain**, University of Manchester
- **Dr Aixin Cheng**, University of Manchester
- **Pinyuan Tian**, University of Manchester
- **Dr Heulyn Jones**, University of Strathclyde

Safety Hub

- **Dr Ioannis Bantounas**, University of Manchester
- **Dr Mike Barrow**, University of Liverpool
- **Dr Joan Comenge**, University of Liverpool
- **Dr John Connell**, University College London
- **Dr Marie Held**, University of Liverpool

- **Dr Inna Linnik**, University of Manchester
- **Dr Stephen Patrick**, University College London
- **Dr Parisa Ranjzad**, University of Manchester
- **Dr Jack Sharkey**, University of Liverpool
- **Dr Philip Starkey Lewis**, University of Edinburgh
- **Dr Dhifaf Jasim**, University of Manchester
- **Dr Yichao Yu**, University College London
- **Dr Fang Zhang**, University of Liverpool
- **Dr Rashida Lathan**, University of Glasgow
- **Dr Arthur Taylor**, University of Liverpool
- **Nathalie De Bois Brillant**, PhD Student, University of Liverpool
- ***Oihane Fragueiro,*** PhD Student, University of Liverpool
- **Ilaria Santeramo**, PhD Student, University of Liverpool
- **Lauren Scarfe**, PhD Student, University of Liverpool
- **Joseph Zeguer**, PhD Student, University of Liverpool

Acellular Hub

- **Ms Mahetab Amer**, University of Nottingham
- **Mr Abdulrahman Baki**, University of Nottingham
- **Dr Defogail Delcassian**, University of Nottingham
- **Dr Deepak Kumar**, University of Manchester
- **Dr Hareklea Markides**, Keele University
- **Dr Jane McLaren**, University of Nottingham
- **Dr Ben Pierce**, Imperial College (Research Co-Ordinator)
- **Dr Jenny Puetzer**, Imperial College
- **Dr Omar Qutachi**, University of Nottingham
- **Dr Robin Rumney**, University of Southampton
- **Dr Jean-Philippe St-Pierre**, Imperial College
- **Dr Lalitha Thiagarajan**, University of Nottingham
- **Dr Emma Wright**, University of Nottingham
- **Dr Scarlett Xue**, University of Nottingham

Immunomodulation Hub

- **Dr Raul Elgueta**, King's College London
- **Dr Giorgia Fanelli**, King's College London
- **Dr Matthias Friedrich**, University of Oxford
- **Dr Peter Gardner**, University College London
- **Dr Helen Marshall**, Newcastle University
- **Dr Jasmine Penny**, University of Birmingham
- **Dr Susanne Sattler**, Imperial College London
- **Dr Fang Xiao**, King's College London

Annex 4

UKRMP Hub publications

PSCP Hub

- *Are Stem Cell-Based Therapies for Parkinson's Disease Ready for the Clinic in 2016?* Barker RA, Parmar M, Kirkeby A, Björklund A, Thompson L, Brundin P. J Parkinsons Dis. 2016;6(1):57-63. doi: 10.3233/JPD-160798.
- *Autophagic response to cell culture stress in pluripotent stem cells.* Gregory S, Swamy S, Hewitt Z, Wood A, Weightman R, Moore H. Biochem Biophys Res Commun. 2016 May 6;473(3):758-63. doi: 10.1016/j.bbrc.2015.09.080. Epub 2015 Sep 15
- *Setting Global Standards for Stem Cell Research and Clinical Translation: The 2016 ISSCR Guidelines* Daley GQ, Hyun I, Apperley JF, Barker RA, Benvenisty N, Bredenoord AL, et. al., Stem Cell Reports. 2016 Jun 14;6(6):787-97. doi: 10.1016/j.stemcr.2016.05.001. Epub 2016 May 12.
- *Making it personal: the prospects for autologous pluripotent stem cell-derived therapies.* Blair NF, Barker RA. Regen Med. 2016 Jul;11(5):423-5. doi: 10.2217/rme-2016-0057. Epub 2016 Jun 27.
- *Comparability: manufacturing, characterization and controls, report of a UK Regenerative Medicine Platform Pluripotent Stem Cell Platform Workshop, Trinity Hall, Cambridge*, 14-15 September 2015. Williams DJ, Archer R, Archibald P, Bantounas I, Baptista R, Barker R, et. al., Regen Med. 2016 Jul;11(5):483-92. doi: 10.2217/rme-2016-0053. Epub 2016 Jul 12.

Niche Hub

- *Pluripotent stem cell derived hepatocytes: using materials to define cellular differentiation and tissue engineering.* Lucendo-Villarin B, Rashidi H, Cameron K, Hay DC. J. Mater. Chem. B, 2016,4, 3433-3442. DOI: 10.1039/C6TB00331A Epub. 15 Apr 2016.
- *Reducing Hepatocyte Injury and Necrosis in Response to Paracetamol Using Noncoding RNAs.* Szkolnicka D, Lucendo-Villarin B, Moore JK, Simpson KJ, Forbes SJ, Hay DC. Stem Cells Transl Med 2016 Jun;5(6):764-72. doi: 10.5966/sctm.2015-0117. Epub 2016 Apr 7.
- *Concise Review: Advances in Generating Hepatocytes from Pluripotent Stem Cells for Translational Medicine.* Szkolnicka D, Hay DC. Stem Cells. 2016 Jun;34(6):1421-6. doi: 10.1002/stem.2368. Epub 2016 Apr 22.
- *Fluid shear stress modulation of hepatocyte-like cell function.* Rashidi H, Alhaque S, Szkolnicka D, Flint O, Hay DC. Arch Toxicol. 2016 Jul;90(7): 1757-61. doi: 10.1007/s00204-016-1689-8. Epub 2016 Mar 15.
- *Mass production of stem cell derived human hepatocytes for experimental medicine.* Wang Y, Hay DC. Expert Rev Gastroenterol Hepatol. 2016 Jul; 10(7):769-71. doi: 10.1080/17474124.2016.1182862. Epub 2016 May 9.
- *A Novel Automated High-Content Analysis Workflow Capturing Cell Population Dynamics from Induced Pluripotent Stem Cell Live Imaging Data.* Kerz M, Folarin A, Meleckyte R, Watt FM, Dobson RJ, Danovi D. J Biomol Screen. 2016 Jun 2. pii: 1087057116652064. [Epub ahead of print]
- *Quantitative multi-image analysis for biomedical Raman spectroscopic imaging.* Hedegaard MA, Bergholt MS, Stevens MM. J Biophotonics. 2016 May;9(5):542-50. doi: 10.1002/jbio.201500238. Epub 2016 Feb 2.
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- *Serum-Free Directed Differentiation of Human Embryonic Stem Cells to Hepatocytes.* Cameron K, Lucendo-Villarin B, Szkolnicka D, Hay DC. Methods Mol Biol. 2015;1250:105-11. doi: 10.1007/978-1-4939-2074-7_7.
- *CSF1 Restores Innate Immunity After Liver Injury in Mice and Serum Levels Indicate Outcomes of Patients With Acute Liver Failure.* Stutchfield BM, Antoine DJ, Mackinnon AC, Gow DJ, Bain CC, Hawley CA et al. Gastroenterology. 2015 Dec;149(7): 1896-1909.e14. doi: 10.1053/j.gastro.2015.08.053. Epub 2015 Sep 5.
- *CNS Myelin Sheath Lengths are an Intrinsic Property of Oligodendrocytes.* Bechler ME, Byrne L, Ffrench-Constant C. Curr Biol. 2015 Sep 21;25(18):2411-6. doi: 10.1016/j.cub.2015.07.056. Epub 2015 Aug 27.

Patent Filings:

- A point of care platform for acute liver injury (Patent number GB 1512219.5); - Dear Lab
- Recombinant laminins drive the differentiation and self-organisation of hESC-derived hepatocytes. (Patent number 62/248,389) – Hay Lab

Safety Hub

- *Imaging technologies for monitoring the safety, efficacy and mechanisms of action of cell-based regenerative medicine therapies in models of kidney disease.* Sharkey J, Scarfe L, Santeramo I, Garcia-Finana M, Park BK, Poptani H, Wilm B, Taylor A, Murray P. Eur J Pharmacol. 2016 Jul 1. pii: S0014-2999(16)30426-5. doi: 10.1016/j.ejphar.2016.06.056.
- *Preventing Plasmon Coupling between Gold Nanorods Improves the Sensitivity of Photoacoustic Detection of Labeled Stem Cells in Vivo.* Comenge J, Fragueiro O, Sharkey J, Taylor A, Held M, Burton NC, Park BK, Wilm B, Murray P, Brust M, Lévy R. ACS Nano. 2016 Jun 20. [Epub ahead of print].
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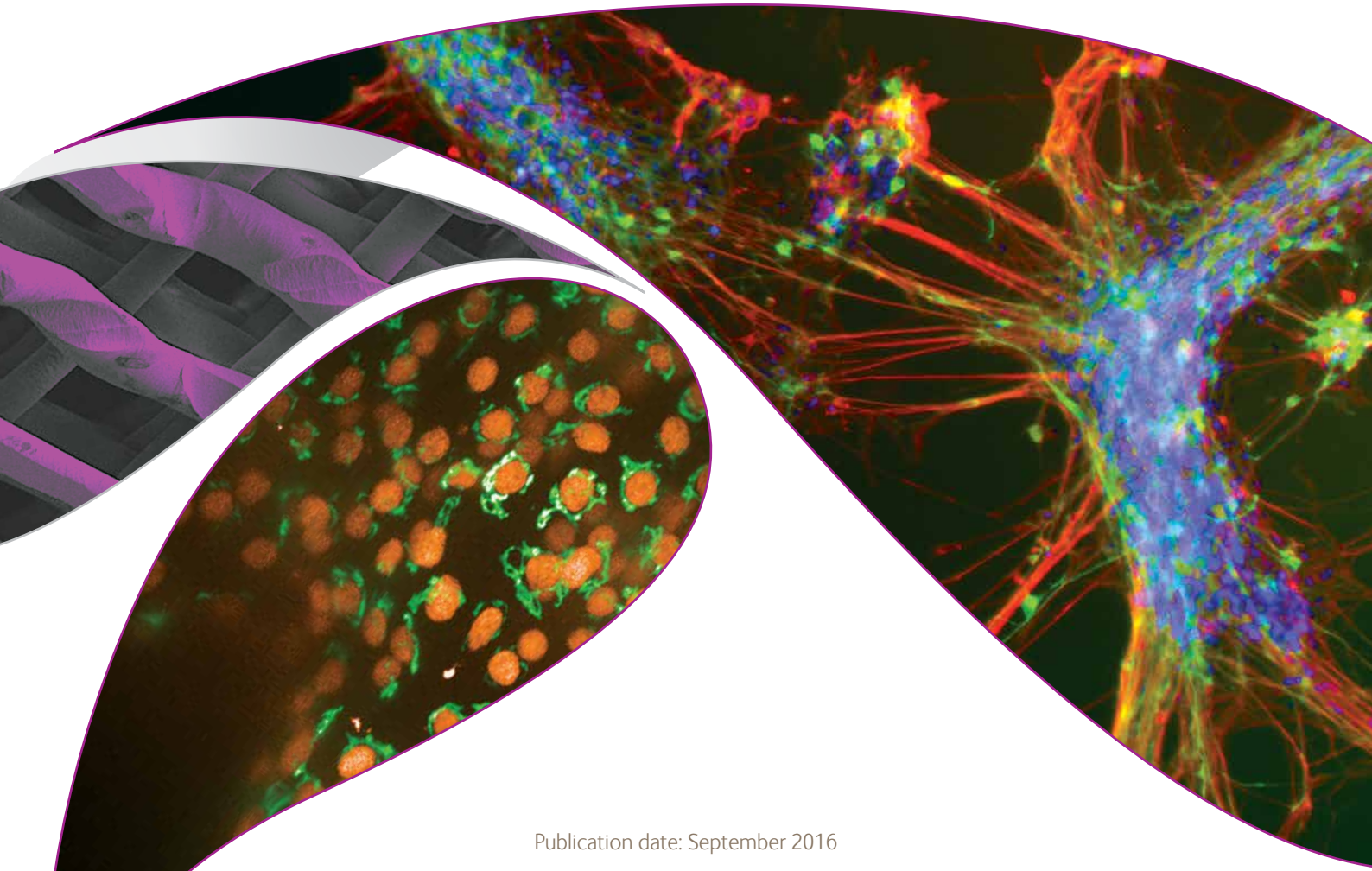

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