National Centre
for the Replacement
Refinement & Reduction
of Animals in Research

UK Regenerative
Medicine Platform

Workshop report:

Applying human cell and regenerative medicine technologies to efficacy and safety testing of new drugs

Workshop hosted by the NC3Rs, UK Regenerative Medicine Platforms, Stevenage Bioscience Catalyst, NAT SIG and Innovate UK

Workshop: 13 April 2015
Report published: 17 December 2015

Pioneering Better Science

Introduction

Cutting-edge cell and tissue based technologies, such as those used in regenerative medicine, offer a tantalising prospect to transform drug discovery. However, their capacity to be applied directly to drug discovery and add value to the industry varies depending on whether the specific technology is ready for scale-up and the complexity of modelling the specific organ system. The NC3Rs, in collaboration with the UK Regenerative Medicine Hubs, Stevenage Bioscience Catalyst, Innovate UK and the Non-Animal Technologies Special Interest Group[1] (NAT SIG) held a workshop to explore the areas with the most promise and identify the next steps for commercialisation, scale-up, increased uptake and integration into drug discovery and development processes.

The scene for the workshop was set by the presentation of industry perspectives on some of the challenges faced in drug discovery and development that may be tackled by the use of cell and tissue-based technologies. Four areas were showcased which currently rely on *in vivo* models (i) biologic development, (ii) cardiovascular toxicity risks for small molecules, (iii) current approaches for modelling kidney toxicity and (iv) immune reactions caused by cell and gene therapies.

Biologic development:

Examples of the types of toxicities observed with large molecules such as monoclonal antibodies were demonstrated using real-life case studies from the pharmaceutical and biotechnology industry. These included sinusoidal dilation in the liver, vascular neoplasms in the skin, heart and lung which are suggestive of pulmonary hypertension, increase in mean arterial pressure and other electrocardiogram (ECG) waveform abnormalities. The most promising opportunity identified for the application of cell and tissue-based technologies in biologics development was 3D liver technology which could potentially have been useful to characterise sinusoidal dilation across species, explore the mechanism of toxicity and develop potential biomarkers for use in the clinic.

Cardiovascular toxicity:

The objective of preclinical studies to detect risk as early as possible, ideally *in vitro*, was described in company case studies on cardiovascular toxicity. Examples of success stories in this area demonstrated how the hERG assay has been used to predict TdP risk and more recently how an iPS cardiomyocyte screen has been developed to detect risk of drug-induced decreases in cardiac contractility in early discovery. The ideal scenario in drug discovery would be to uncover both functional and structural cardiac changes in screens that have high enough throughput and low enough cost to enable their positioning in early discovery. The major short-falls of the currently available systems are the inability to detect effects on (i) cardiac structure (usually only observed after chronic dosing) and (ii) cardiovascular haemodynamics.

[1] The NAT SIG is delivered by the NC3Rs in collaboration with the Knowledge Transfer Network and Innovate UK

Introduction

Modelling kidney disease:

There are a variety of *in vivo* models used in drug development to assess efficacy of candidate drugs to treat kidney disease. These include ischaemia reperfusion injury, unilateral uretal obstruction and adriamycin-induced nephropathy models, all of which lead to progressive renal fibrosis. Modelling particular aspects of kidney disease, such as tubular necrosis, tubular degeneration, tubular dilation and the resulting nephropathy can be difficult in cell or tissue-based assays. However, *in vitro* models are still extremely valuable in the evaluation of drug toxicity and regenerative medicine therapies. For instance, 3D models using *ex vivo* kidney rudiments can be used to determine which cell types have the potential to generate specialised renal cells which is important in assessing efficacy of potential regenerative medicine therapies. Participants at the workshop discussed how a lack of understanding of recent developments in some cell and tissue-based models of kidney disease can contribute to a lack of confidence in the models. This could be addressed by providing incentives to increase and accelerate their use which will enable greater understanding of their capabilities and limitations.

Immune reactions caused by cell and gene therapies:

A further area where cell and tissue based technologies have potential is in assessing immune system reactions. This is important for gene therapy products for a number of reasons. The immune system could be (i) an intended target for treatment of immune system disorders, (ii) used or engaged to treat diseases in other organs (e.g. cancer) and (iii) be triggered unintentionally causing immune-mediated adverse reactions (e.g. cytokine storm); all of which need to be understood prior to clinical trials. At the workshop the focus was on the potential of cell and tissue-based systems to aid in the prediction of immunogenicity which may be variable due to the vector, the expressed protein, disease background, patient age and the route of administration and/or site of gene expression.

Industry landscape and exploiting new technologies:

Despite the challenges described there is support and desire from industry to use cell and tissue-based technologies. Industry are keen to see some test compounds run through the different systems in development so the discussion can progress towards what the data looks like, interpretation of the data in practice, which scientific questions or business decisions the assays could be used for and the potential limitations of the assays. One of the current challenges for the technology developers is access to compounds and advice on the type of compounds that should be used to test the systems. To accelerate this, collaborative projects will be essential to ensure there are sufficient numbers and diversity of compounds to trial the technologies and that the resource required to fund such exploratory projects is spread across interested parties. The workshop explored how the scientific community including academic scientists, industry scientists, business developers and funders, could work together to create such collaborations in the UK.

Horizon scanning

The UK Regenerative Medicine Platform Hubs provide a resource for collaborators looking to access research and methodologies in stem cell biology, tissue engineering methods and analysis. There are five Hubs that presented an overview of their work at the workshop which is summarised below:

Acellular technologies (http://www.ukrmp.org.uk/hubs/acellular/)

The acellular Hub develops platform technologies that use materials and drugs to orchestrate cells to regenerate tissue. Relevant research areas include 3D printing of cells and materials to automate the production of precise *in vitro* cultures, electrospinning to create fibrous scaffolds for epithelial cell culture, patterning of cells in tubular tissue structures and peptide hydrogels to control local mechanical properties around cell populations.

Safety and efficacy, focusing on imaging technologies (http://www.ukrmp.org.uk/hubs/safety/)

The overall aim of this Hub is to enable clinical translation of safe effective regenerative medicine therapies through integration of the therapeutic safety expertise of the MRC Centre for Drug Safety Science. The Hub is developing and applying cutting edge imaging technologies, nanochemistry, stem cell biology and clinical methodologies. Imaging technologies in particular, for example light sheet microscopy, could contribute to the development, evaluation and analysis of complex 3D tissue mimics for drug testing.

Engineering and exploiting the stem cell niche (http://www.ukrmp.org.uk/hubs/niche/)

This Hub is identifying key factors that promote adult and pluripotent stem cell differentiation, molecular targets to direct stem cells to promote endogenous repair and to enhance the longevity and function of transplanted cells. The focus on differentiation of stem cells has clear implications for the development of new human-based technologies for drug efficacy and safety screening.

Immunomodulation (http://www.ukrmp.org.uk/hubs/immunomodulation/)

The focus of the immunomodulation Hub is to discover how to harness the immune system for improved outcomes in regenerative medicine. Some of the clinical targets of this Hub include: improved efficacy of photoreceptor cell therapy to treat blindness; improved repair of damaged heart tissue; improved survival and functionality of transplanted hepatocytes as an alternative to liver transplantation; and understanding how inflammation affects tissue repair.

Cell behaviour, differentiation and manufacturing (http://www.ukrmp.org.uk/hubs/pscp/)

This Hub aims to optimise processes for consistent, scalable stem cell manufacturing that minimise the appearance of genetic and epigenetic variants, and meet the requirements of clinicians, regulatory authorities and industry for safe and cost-effective applications.

Key highlights from the breakout sessions

The breakout sessions were split into two parts. The first was to identify the scientific and technological knowledge gaps which limit the application of cell and tissue-based models in the areas presented by industry in the introductory session: biologics, cardiovascular toxicity, kidney disease and immune system related safety issues in gene therapy. The second was to discuss: frameworks for future investment in platform development and commercialisation, priority research areas and infrastructure and collaboration models.

The main findings from the breakout sessions are grouped and described below.

Identifying the next tractable problems:

Further investment into the current funding mechanisms from the NC3Rs, Innovate UK, the Research Councils and other funding bodies which are specifically focused on bringing together interdisciplinary teams to solve problems using cell and tissue-based technologies was identified as critical to enable the UK to capitalise on its unique strengths in this area. Scientists and technologists across sectors will need to identify realistic first targets for *in vitro* models where progress can be made that will impact on the quality of data generated by cell and tissue-based methods. For example, in the breakout session focusing on the kidney it was proposed that fibrosis was a realistic target that would be likely to result in a functional cell-based model within five years. The cardiovascular breakout group discussed how 17-20% of drug attrition from cardiovascular liability is due to structural toxicity, detection of which is not currently possible using *in silico* or *in vitro* systems. As a consequence, animal use remains high. Development of new biomarkers of cardiotoxicity was proposed as a priority to enable better predictivity of clinical outcome and reduction of animal use. Pharmaceutical industry scientists also highlighted the need for assays that reported on acute, chronic and multi-dose cardiotoxicity.

Making differentiated cells from pluripotent stem cell sources:

A common opportunity which spans many different cell and tissue types is the use of stem cell technologies to make cells where primary cells cannot be expanded. For cardiovascular toxicity, production of iPS cardiomyocytes is now relatively routine in academic and commercial laboratories. While maturation of the iPS cardiomyocytes remains an issue, there are growing numbers of examples where these cells have been useful to both industry and academia to identify beneficial or harmful drugs. A number of global initiatives, including an NC3Rs CRACK IT Challenge (InPulse), are currently addressing issues around maturity and contractility.

Therefore, the next step should be to look beyond cardiomyocyte production and maturity into future programmes which construct "intelligent" tissues combining iPS cardiomyocytes, endothelial cells, smooth muscle and fibroblasts. The technological leap into this next stage is likely to be the inclusion of fluidics, imaging and the ability to detect secreted biomarkers. Gene targeted iPS lines that report on cell stress, proliferation, energy and hypertrophy would also be beneficial. Thinking even further into the future, opportunities to build in *in silico*, machine learning and personalised approaches (through cells derived from patients with specific diseases) were identified.

Key highlights from the breakout sessions

For cell and tissue-based technologies to be applied to industry, the constructs will need to be viable for a minimum of two weeks spanning up to three months to enable the longer term, multi-dose studies for chronic testing required by the industry. The format would be at least 96-well to allow sufficient throughput in the 10-1000 compound range. Cost of the platform is a key consideration.

Adding value to *in vitro* models:

The latest stem cell and tissue engineering techniques are generating possibilities to improve tissue architecture and complexity in *in vitro* models especially if combined with imaging techniques that enable them to be analysed live and in real time. This complexity will enhance currently available models to better mimic tissue functions *in vivo*. There were opportunities identified such as interfacing tissue models with immune system cells and generating *in vivo* tissue outputs from *in vitro* models, for example, cardiac muscle models that include structural changes on exposure to drugs that match the changes seen in animal and patient tissues. Clinically-focused activities to improve scale up and manufacture of complex tissue products could help to minimise the cost of these more complex *in vitro* models and allow automation of the production of identical tissue models.

Validation through open innovation and collaboration

Participants at the workshop agreed that the term 'validation' in the context of advancing cell and tissue-based technologies needed to be distinguished from the formal validation process to replace *in vivo* studies at a regulatory level. A more realistic and positive interpretation of 'validation' refers to generating a threshold of scientific evidence to demonstrate that a specific technology is useful in predicting human efficacy or toxicology during compound development.

In this context there are two types of evaluation of cell and tissue-based technologies. One is the technological evaluation of the system to demonstrate that it functions as expected over a period of time, for example by producing specific markers that are indicative of function. This evaluation should be integrated throughout the development of the technology and also during the scale up and manufacturing process.

The other is predictive evaluation which should assess whether the model predicts human outcome in the clinic or is reliable enough to make sound decisions on compound progression. A key theme that resonated with all participants was that prior to any larger scale validation exercise there should be significant discussion amongst the expert community to determine the precise scientific question the new technology could be applied to. This would lead to more thoughtful and intelligent use of the tools on a case-by-case basis and improved understanding of what was needed by the end-user (e.g. medium vs high throughput) which in turn would lead to higher likelihood of success. One of the key needs that catalysed this discussion was the necessity to identify biomarkers predictive of clinical responses to stem cell therapies. The disease status of individual patient is likely to produce significant changes to the cells administered. Therefore, the molecular characterisation (using biomarkers) of the patient at the time of treatment will be important to provide the crucial information needed to improve stratification.

Key highlights from the breakout sessions

The coordination of available methods, knowledge, data and compounds from companies would be invaluable in progressing cell and tissue-based technologies. A stepwise approach was suggested where initially the community could share information and experience on methods and technologies that are currently in use. This foundation could lead to sharing of actual technologies and test-systems between laboratories and companies.

A clear barrier to validation currently is the lack of relevant and accepted tool/compound sets. As a first step it was suggested that the community could develop a list of compounds relevant to each organ or tissue that would provide initial decision making data on whether the technology was promising or not. Initially this would be for a set of priority organs such as liver, heart, and lung, and could be based on publically available information such as compounds listed in CRACK IT Challenges and the AstraZeneca clinical compound bank. Scientifically relevant compounds would need to be carefully selected. For example, for cardiotoxicity the list would need to include compounds that cause structural necrosis. These lists would evolve as momentum, input and experience increased.

Sharing data on compounds, particularly failed compounds and also making compounds available for retrospective studies would be the next step and should be supported by an assigned honest broker organisation that could collate, anonomyse and analyse the data. Comparative data across species and clinical data would also be needed. In some cases, validation may involve the co-development of an animal *in vitro* system to increase confidence and tease out *in vitro* and *in vivo* differences from species differences.

A library which made the compounds on the lists available to technology developers has the potential to significantly accelerate progress and validation. Gaining access to such resources has traditionally been very difficult; however, the momentum around these technologies and the involvement of the right partners may create the critical mass necessary for change. The infrastructure and funding for a safe-harbour of compounds model could potentially use the European Lead Factory (IMI) or the Royal Society of Chemistry as an example.

A mechanism to enable fluid and rapid movement of researchers and tools between organisations, sectors, disciplines and networks through secondments was identified as also being extremely valuable to accelerate validation of new technologies.

Capitalising on the world-leading UK environment:

The UK has an exceptional and unique environment to capitalise on the opportunities that were identified at the workshop. The existing world-leading Regenerative Medicine Platforms Hubs, the Integrative Knowledge Centres (EPSRC) and Catapults (e.g. Precision Medicine, Medicines Technologies and Regenerative Medicine Catapults) with areas of overlapping interests give the UK an internationally competitive base that could be strengthened specifically in the application of cell and tissue-based technologies. Such an approach would involve intelligent prioritising of efforts not to compete with the EU and US but capitalising on the UK's strengths.

Key highlights from the breakout sessions

In addition to the scientific centres of excellence, the UK also has well-established collaborative activities between the NC3Rs and Innovate UK, and the NC3Rs and the MHRA. The UK regulatory environment is progressive and open to non-standard approaches to predicting human efficacy and safety through strong links with scientists and other organisations in the UK. One of the key aims of these collaborations is to accelerate evidence-based changes in the use of non-animal approaches such as cell and tissue-based technologies. Key opinion leaders, the Research Councils and other departments in Government have collaborated with Innovate UK and the NC3Rs on a roadmap for developing and commercialising non-animal technologies which shows the UK commitment and clear strategic vision in this area.

To maximise the potential of this powerful and developing environment key areas for future strategy were identified during the workshop. These included increased precompetitive collaboration across sectors (including more data-sharing), better outreach of the existing organisations (e.g. Regenerative Medicine Platforms) to chemical and consumer product companies and focused coordination to bring all the disciplines together specifically for the applications related to drug and chemical development.

The ability to use stem cell technologies and regenerative medicine to develop *in vitro* tissues that replicate key functions of human tissues and organs creates new opportunities to transform the discovery of drugs and advanced medicines. The UK has critical mass and proven excellence in science and translation in these fields. A major opportunity exists to establish the UK as the leading country for solving industry focused problems that accelerate drug development by enhancing non-animal technologies.

Conclusions and recommendations:

Recommendations from this workshop focus on better defining the problems that can be solved together by communities by forming long-term partnerships between large companies, SMEs, universities and charities.

Identifying the next tractable problems:

- Coordinate the UK community to work in partnership to define focussed, translatable and tractable problems that can be solved using cell and tissue-based technologies.
- Demonstrate successful examples of viable cell and tissue-based technologies to highlight scientific and commercial feasibility and industrial 'pull-through'.
- Expand collaborative programmes for challenge-led innovation and strategic funding (e.g. CRACK IT).

Making differentiated cells from pluripotent stem cell sources:

- Identify key target cells and potential stem cell source with high likelihood of success, for example, specialised renal cells (such as proximal tubule cells and podocytes), hepatocytes, and cardiomyocytes.
- Focus on specific patient groups where tissue could more easily be accessed, collected and banked.
- Prioritise the development of identified cell lines and tissues in a coordinated UK science and business strategy between funding bodies, Research Councils, existing Catapults, Innovate UK, the NAT SIG and the NC3Rs.

Adding value to *in vitro* models:

- Increased focused activity to nurture long-term, cross-discipline (industry/SME/academia) collaboration will be critical to achieve a step-change in scale-up and manufacture of cell and tissue-based technologies.
- Inclusion of structural and functional endpoints of cell and tissue-based technologies and a robust strategy which enables early go/no go decisions on technological and manufacturing feasibility will maximise the benefits of such a focused activity.

Validation through open innovation and collaboration

- Establish an honest broker to work with technology developers and end-users to develop a compound library (including associated *in vivo* data and mechanistic/pharmacology information where possible), a resource for data sharing and a partnering hub.
- The NC3Rs has a first rate reputation for fostering partnering and data sharing activities, and also manages the NAT SIG. It would therefore be uniquely placed to expand this role.
- Explore the use of other models to increase openness and data sharing (e.g. the Open Science Framework).
- Make use of an existing or create a new facility/bank to store a compound library for testing cell and tissue-based technologies.

Conclusions and recommendations:

Capitalising on the world-leading UK environment:

- Continued and increased collaboration between funders for example the NC3Rs, Innovate UK, and the Research Councils to nurture long-term partnerships between leading UK scientists and centres of excellence.
- The importance of the UK research base in contributing to innovation and entrepreneurship in the area of cell and tissue-based technologies should be recognised and supported by Government as described in the Witty report as 'Arrow Projects'. This will drive forward the technological ideas from universities (at the tip of the arrow) into real commercial prospects (the arrowhead of related economic activity).
- Ensure responsive and flexible access to resource and funding to take ideas forward from research to commercialisation and foster the technology pipeline at all stages. Improving pump-prime funding and reducing complexity in policy support mechanisms for research and innovation has also been identified as essential to UK science and business in the Dowling review.
- Maximise the potential of the existing UK environment. Engage and coordinate SMEs, large pharma, academia, contract research organisations, NAT SIG, Catapults, funding bodies and research charities to tackle the next tractable problems specifically where cell and tissue-based technologies can have a significant impact.

About the hosts

National Centre for the Replacement Refinement & Reduction of Animals in Research

The NC3Rs is a UK-based scientific organisation dedicated to replacing, refining and reducing the use of animals in research and testing (the 3Rs). We collaborate with scientists and organisations from across the life sciences sector, nationally and internationally, including universities, the pharmaceutical, chemical and consumer products industries, other research funders, and regulatory authorities. We support the commitment of the scientific community to the 3Rs by funding research and early career development, supporting open innovation and the commercialisation of 3Rs technologies, and stimulating changes in policy, regulations and practice. For further information about NC3Rs activities and programmes see www.nc3rs.org.uk.

The Non-Animal Technologies Special Interest Group (NAT SIG) connects the research and business communities in the development and application of novel technologies that have the potential to transform business and improve product development across a range of industries, and reduce reliance on animal models. It provides a platform for showcasing the latest news, events and funding relevant to the non-animal technology community, as well as hosting networking events and workshops to meet the recommendations set out in the non-animal technologies roadmap. The NAT SIG is a partnership between the NC3Rs and Innovate UK. For further information see https://connect.innovateuk.org/group/non-animal-technologies.

UK Regenerative Medicine Platform

The UK Regenerative Medicine Platform (UKRMP) addresses the technical and scientific challenges associated with translating promising scientific discoveries in regenerative medicine towards clinical impact and seeks to provide a world-leading programme to promote the development of regenerative therapies. Central to the Platform are five interdisciplinary and complementary research Hubs with the necessary critical mass to address key translational challenges and provide new tools, protocols and resources with broad applicability that can be utilized by other UK research groups in academia and industry. It is funded as a single joint programme, through the BBSRC, EPSRC and MRC. For further information see www.ukrmp.org.uk.

STEVENAGE BIOSCIENCE CATALYST
Open Innovation Campus

Stevenage Bioscience Catalyst is the UK's first open innovation bioscience campus, pioneering a unique culture to drive early stage bioscience technology and company development, and building a thriving community. It is backed by £38m of funding from its founding partners – GlaxoSmithKline, the Wellcome Trust, the Department for Business, Innovation and Skills, Innovate UK and the former East of England Development Agency. Consisting of an Incubator, an Accelerator and a Hub, covering 60,000 sq ft of laboratory, office and networking space, the independent facility houses a range of companies, from virtual and start-up firms to those which are more established, as well as other organisations. Co-located with GlaxoSmithKline on the Stevenage site, Stevenage Bioscience Catalyst is in the unique position of operating in proximity to the expertise and resources of a major pharmaceutical company, close to both London and Cambridge. For further information see www.stevenagecatalyst.com.