From Bedside to Bench: Preclinical Target Validation using Patient Derived Cells

The SGC, its pharmaceutical company partners, clinicians, hospitals and disease foundations launch collaborative open source pre-clinical translational medicine studies

The problem: Lack of predictability

Although the number of annual drug approvals is trending upward, the number of 'first in class' therapies has remained relatively constant, often less than 10 per year. Despite the recent successes, the number of new approved drugs are still below that observed in the mid-nineties. For such new medicines for 'pioneer targets', the level of attrition in Phase 2 proof-of-concept clinical studies remains the biggest hurdle [**REF: BUNNAGE**], in large part because the target-disease associations derived from the currently dominant cell or animal pre-clinical models of disease most often do not translate into clinical efficacy. It is increasingly appreciated that the use of disease models based on human samples will be critical both to increase our understanding of pathophysiology and improve the productivity of the sector. However, securing regular access to well-annotated samples from patients is challenging to organize, raises ethical issues and requires new organizational models between universities, hospitals, disease foundations and the private sector. We propose that these issues can most easily be addressed by creating an open source partnership.

The scientific need: predictive human tissue and primary cell based assays

Preclinical studies with conventional cell lines and animal models of disease have often proven unable to predict drug efficacy in human trials. In some cases, for example *sepsis*, hundreds of compounds are effective in animals, but none have proven efficacious in humans [**REF: MARSHALL**].

The discovery of anti-TNF therapy of RA provides an example of where the use of novel human tissue helped define and validate a target, and also of the need to mimic the disease condition well. In the mid 1980's, synovial cultures from patients with RA were based on fibroblast-like synoviocytes, which are easy to culture but only comprise 10% of the total cell population. The remaining inflammatory and immune cells, which need specialized conditions, including hypoxia and growth factors, survive poorly in crude cell cultures and were discarded. Only when new culture methods were developed that enabled 90% of the total cells originating from the diseased joint to survive for 5-6 days, was it possible to provide the first convincing evidence for the importance of TNF in joint inflammation, which thereafter was rapidly confirmed in animal models and the proof of principle trials **[REF: BRENNAN].**

Other inflammation targets were validated in work based on patient-derived cells. IL-17 was identified as an important target in autoimmunity based on work with synoviocytes and synovial explants from patients with *rheumatoid arthritis* (RA). The idea was supported by the identification of Th17 cells, primarily in mouse models. However, the mouse models provided confusing data and it was not possible to predict the specific diseases in which an anti-IL-17 therapy might be effective. Human genetic association studies eventually pointed to links with *psoriasis*, *psoriatic arthritis* and *ankylosing spondylitis*, and now multiple studies have now shown that anti-IL-17 targeted therapies are clinically effective. **[REF: TBC]**

These and other examples highlight the advantages of using patient-derived tissue. But the discovery of anti-TNF therapy also provides two other lessons. *First,* success derived not only from the use of human samples by expert clinicians, but also from the use of excellent reagents, in this case industry-derived antibodies to important mediators. The *second* lesson was that the speed by which the discoveries were translated into medicines was in large part due to the openness of the scientists. This pre-clinical work was published immediately and in addition, the clinical finding that patients dramatically responded to anti-TNF antibody was disclosed more than a year before publication.

Our goal is to learn from these and other examples and build a scalable new organization to recapitulate their successes.

The organizational solution: An open partnership between researchers in academia and industry, hospitals and patient groups

Our aim is to identify and validate pioneer targets by profiling the highest quality chemical and antibody tools in the highest quality assays derived from patient cells or tissues. The main challenges in accomplishing this is not conceptual, but rather operational and organizational. Operational because selective, drug-like molecules or specific antibodies are challenging to discover and because large well-annotated patient cohorts are rare and difficult to access. Organizational because all the necessary ingredients are rarely found in the same institution: industry usually has the most experience in the design and development of new chemical agents or antibodies; the clinical academic community cares for the patients and provide deep disease expertise; and the academic research community customarily provides the molecular and technological insights necessary for mechanistic studies.

Clearly, the ideal way to discover better drug targets is to develop an organizational model that allows academics, clinicians, foundations and drug hunters to collaborate seamlessly; this is not easily achieved in the current biomedical system. Accordingly, we are establishing an *open source target discovery partnership* in which pharmaceutical companies collaborate with academic scientists and clinicians to generate high quality chemical and antibody probes for unprecedented targets and profile them in patient-derived cells from selected diseases. The targets will be selected based on novelty and tractability, with initial emphasis on epigenetics and protein kinases, and with later emphasis on targets suggested by human genetic studies. The initial diseases and the participating centers will be selected based on *i)* the availability of blood and tissue on an ongoing basis, *ii)* the degree of characterization of the patient cohort, *iii)* the willingness to share reagents, data and knowledge without restriction, and *iv)* the willingness to host a cell assay team that works to industry standard quality metrics.

The commitment to open source and data sharing is a key feature of our partnership, and is expected to accelerate the science, make the data generation more transparent and thus more reproducible, and reduce significantly the transactional costs associated with multi-institutional, multi-national and multi-sector collaborations. It is also expected to significantly alleviate ethical concerns that might arise when commercial and scientific interests are juxtaposed with patient samples.

Progress

The partnership builds from the SGC, a public-private partnership that has proven able to generate and disseminate high quality research tools (www.thesgc.org). In our new open source target discovery partnership, these and other high quality tools will be distributed to an organized network of hospital-affiliated laboratories around the world, where they will be profiled using state-of-the-art and clinically relevant assays based on patient derived cells.

In Europe, as part of a new project supported by the Innovative Medicines Initiative, the partnership will focus on diseases within **inflammation and auto-immunity**, including *fibrosis, ankylosing spondylitis, lupus and myositis*. Tissues and cells will be collected from well-characterized and managed patient cohorts in Oxford and at the Karolinska University hospital. Using cells from patients, we will generate robust, clinically meaningful assays, and use those to profile molecularly targeted probes using phenotypic and biomarker readouts (such as cytokine profiles), linking novel targets to important new indications. Dedicated assay teams will be put in place; this will best ensure quality and reproducibility. In addition to the assays already established by our clinical partners*, e.g.* for *myositis* and *fibrosis*, we will also develop innovative new assays with potentially superior translational potential. This may involve more complex culture systems, more closely resembling

human tissue, typically having more than one cell type and in some instances cells embedded in matrix.

Within Canada, we will focus initially on **oncology** with emphasis on cancers that can be cultured *ex vivo*, yet maintain important clinical features of the disease such as histopathology, ability to engraft, and clinical response to standard of care therapeutics. This approach is enabled by recent advances in isolation and culture of cancer stem cells capable of engraftment **[REF: POLLARD]**, 3D cultures of organoids, and cultures grown on materials mimicking the extracellular matrix or with feeder cells that mimic elements of the tumor microenvironment. The use of these techniques makes it feasible to assay a wide panel of samples with the intent to cover, as much as is possible, the heterogeneity observed in the clinic. Responses to probes will be profiled, monitoring features such as cell growth and viability, differentiation/senescence, response to standard of care, and epithelial-mesenchymal transition.

Although formally still to be confirmed in systematic studies, we believe that this approach, which has worked in the past, may better predict efficacy in clinical trials since primary patient cells better represent the genetic and clinical diversity of the human disease. We also believe that, by comparing the same molecular tools across multiple diseases, we will gain tremendous insight into some of the shared molecular mechanisms of subtypes of disease, as well as for those common to cancer and inflammation.

Future directions and a call for new collaborators

The two main objectives of our initiative are to (1) significantly increase our understanding of the molecular basis of cancer and inflammatory disease, as well as in other diseases as our network grows; and (2) identify specific targets for which pharmacological or biotherapeutic modulation ameliorates key disease phenotypes. For some diseases and cell types, such efforts may include the use of induced pluripotent stem cells, for example to generate neuronal cells from somatic cells of patients with neurological disease. Pilot projects are already underway with cells from patients with Rett's syndrome.

Importantly, all projects within the partnership must have an unwavering commitment to open access. No participant will file for patent protection on any result from the cell-based assays and all reagents will be made available to the research community without restriction. For assay results derived from clinical biospecimens, the public will receive aggregated, quality assured and analysed data. Data and cells from individual patients, even though anonymized, cannot be distributed freely due to ethical considerations and associated legislation.

The partnership currently comprises ten pharmaceutical companies and has research sites at the University of Oxford (UK), University of Toronto (Canada), Karolinska Institutet (Sweden), ETH Zürich (Switzerland) and University of Campinas (Brazil). Clinical nodes have been established at the Karolinska University Hospital in Sweden, the Kennedy Institute at the Nuffield Department of Orthopaedics, Rheumatology and Musculoskeletal Sciences in the United Kingdom, as well as at the Hospital for Sick Children, the University Health Network and the Montreal Neurological Institute in Canada. We expect the number of partners to increase as the project progresses.

We also expect that patient organizations will be interested to participate, because our partnership's commitment to open access and its focus on translational studies is completely aligned with their objectives. Indeed, the CHDI Foundation (www.chdifoundation.org) has already joined the partnership to mount an open source search for new drug targets for Huntington's disease. Working together in open seamless collaboration must be better than the status quo.

References

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