Commentary

Organs-on-chips (microphysiological systems): tools to expedite efficacy and toxicity testing in human tissue

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***Experimental Biology and Medicine* 2014; 239: 1073–1077. DOI: 10.1177/1535370214538916**

Introduction

Although basic scientific research has generated methodological and technical innovations rapidly in recent decades, translational science has not kept pace.[1,2] Among the factors contributing to the slower pace of translation is a bottleneck in the drug discovery pipeline.[3,4] This is due in part to the widespread failure of therapeutics in early phase clinical trials, despite promising results from preclinical testing. Improved tools that better predict adverse effects in humans for therapeutics are much needed to reduce the rate of failed trials. Safety, toxicity and efficacy assessment using human tissue as a supplement to animal models prior to clinical testing in humans could reduce clinical trial failure rates,[5] leading to significant savings in lives, time and money.

The Microphysiological Systems (MPS) Program (''organs-on-chips'') (http://www.ncats.nih.gov/research/reengineering/tissue-chip/tissue-chip.html) supports an innovative approach to preclinical toxicity testing on human tissue: development of *in vitro*, three-dimensional organ systems from human cells on bioengineered platforms that mimic *in vivo* tissue architecture and physiological conditions in order to facilitate and accurately monitor key organ-level functions. The platforms incorporate complex factors found *in vivo*, including extracellular scaffolding, three-dimensional structure, cellular interactions (including between different cell types), perfusion, biomechanical stresses (e.g., stretch and shear forces from fluid flow), electrical stimulation of excitable tissue, hormone responses, etc. These features are present in animal models, but some aspects of animal physiology do not accurately represent those of humans.[6] Development of human organ microsystems to bridge the gap between preclinical testing in animals and human clinical trials would be a significant advancement.

History of the Program

A key factor for translational research and drug discovery will be advances in regulatory science. In 2010, the National Institutes of Health (NIH) and the Food and Drug Administration (FDA) co-funded the Advancing Regulatory Sciences Initiative (http://www.nih.gov/news/health/jul2012/ncats-24.htm), a three-year program to accelerate the development and use of new tools, standards and approaches to efficiently develop therapies and to more effectively evaluate product safety, efficacy, and quality. Among the pioneering projects awarded through this program, the Harvard Wyss group engineered a Heart–Lung micromachine for safety and efficacy testing that recapitulates multicellularity of human tissue and mimics the physiology of the human heart and lung (http://wyss.harvard.edu/viewpressrelease/99/wyss-institute-models-a-human-disease-in-an-organonachip-).[7,8]

Following the promise and potential of the heart–lung micromachine, the MPS Program was launched in 2012 by the NIH, the Defense Advanced Research Projects Agency (DARPA), and the FDA (http://www.nih.gov/news/health/jul2012/ncats-24.htm). The MPS Program is a novel five-year partnership to address difficulties in the drug discovery pipeline by providing an alternative approach to traditional *in vitro* cell culture and animal models, thus bridging the gap between preclinical and clinical testing. The MPS Consortium includes staff from NIH, DARPA, FDA, and awardees consisting of scientists, physicians, and engineers with expertise in each of the 10 major organ systems (intestine, liver, central and peripheral nervous system, blood–brain barrier, vascular system, skeletal muscle/innervated motor unit, heart, lung, kidney, and female reproductive system), bioengineering, stem cell biology, cellular and molecular biology, physiology, pharmacology, toxicology, imaging, and fabrication. Concurrent with the immediate goal to develop viable human organ systems is expanded development of bioengineered platforms with viability for at least four weeks. The platforms are engineered to mimic key physiological conditions that support function of each of the organ systems. The platforms also incorporate readout tools for monitoring organ function, including microfluidic sampling and in-line analyzers,

optical and Ca^{2+} imaging, mass spectrometry, contractility measurements and other physiological parameters.

Management of the Program

The long-term goal of the MPS Program is to integrate the 10 major organ modules into a ''human-on-a-chip'' for multi-organ toxicity testing. Maintaining the focus of the multidisciplinary MPS Consortium on this long-term goal necessitates close coordination of investigators' efforts. This is accomplished by a unique approach to management of the program. First, unlike standard research grants, awards were made as milestone-driven cooperative agreements in which government program officials are collaboratively involved with the researchers on each project. Failure to meet milestones can be a basis for stopping a project or establishing steps for course correction. Second, close cooperation between program staff and investigators stimulates progress. Third, frequent updates and regularly scheduled meetings assist with understanding project status relative to the five-year objective, identifying the consortium's current needs, and heading off any potential problems with solutions or course corrections. Fourth, semiannual meetings of the investigators promote timely exchange of data, resources and expertise, and strengthen collaborations. Members of the MPS Consortium can take advantage of the expertise and resources available among their peers to help meet milestones. Early collaborations have been crucial to the goal of integrating organs-on-chips for toxicity screening and drug efficacy.

To date, researchers in the MPS Consortium have progressed rapidly. Work reported in this volume illustrates the progress of the individual research teams, utility of the consortium's collaborative nature, and potential for the MPS Program to reach its primary objective while also generating multiple tools, approaches and potential applications for basic science and medicine. It is also worth mentioning that as a consequence of close and periodic interactions with investigators, NIH made available additional funding via administrative supplements to foster collaborations and the exchange of materials and resources; a repository to induced pluripotent stem cell (iPSC) sources for quality control and characterization; and a library of training and validation compounds.

Partnerships

Multiple Institutes/Centers (I/Cs) across the NIH, led by the National Center for Advancing Translational Sciences (NCATS), comprise the MPS Consortium Project Team. Program Officials from across the NIH provide diverse expertise to address scientific issues related to models of different organ systems, stem cell technology, bioengineering, toxicity, etc. As the MPS Program continues successfully to generate deliverables useful to the consortium and the broader scientific community, it is appropriate (given the relevancy of these projects) that multiple NIH Institutes help sustain MPS momentum by supporting additional spin-off or co-sponsored programs and the NCATS is interested in continuing with them in this endeavor.

Another crucial partnership for the consortium is with the FDA, which has provided regulatory insight since the inception of the MPS Program. This invaluable collaboration provides MPS project investigators with regulatory principles to consider as they design and test their organ systems. Organs-on-chips will go through rigorous tests and comparisons to standard *in vitro* and *in vivo* models (pre-clinical research). Standardization will occur as the chips' reliability and reproducibility are demonstrated further, with the goal of achieving validation and acceptance by regulatory agencies. The FDA's involvement with the MPS Program from its initial stages ensures a solid basis for project development, helping investigators to define appropriate milestones to address crucial regulatory requirements, implementation and acceptance by the FDA.

Importantly, parallel funding of two project teams by DARPA to develop platforms that simultaneously support 10 organ systems is the driving force in the establishment of the human-on-a-chip goal and fosters the collaborative nature of the MPS Program. Platforms developed by the DARPA-supported projects are to be modular and flexible, to allow integration of optimal individual organ systems and support physiologically relevant organ interactions. The goal of multi-organ integration onto a common platform dictates that the most viable, reproducible and physiologically relevant organ systems are selected for integration onto the platform; that optimized common perfusion systems, perfusion medium, pumps, valves and controllers be developed; and that access for testing, imaging, sampling, detection, etc. be engineered into the platforms. It is such program designs that prompted collaboration among MPS, DARPA, and NIH investigators to engineer their platforms in anticipation of integration, and to work toward common requirements, such as platform design and vascular and perfusion systems.

The MPS Project Investigators, the most essential component of the consortium, have done an exemplary job in creating technologies that can mimic human organ function. Platforms engineered by project teams have the potential to change the landscape for preclinical testing of pharmaceuticals and to contribute highly useful tools for biomedical research. In fact, a number of the project groups have already initiated or established collaborations with biotechnology or pharmaceutical companies that are interested in using the MPS platforms to test proprietary compounds. The support of productive public–private partnerships, in conjunction with input from the FDA will accelerate the pace of platform development, refinement, implementation, and regulatory acceptance. The interchange of ideas among basic scientists, the private sector and regulatory agents will accelerate the development of platforms as useful tools and facilitate their availability and delivery to industry and the research community, including pharmaceutical and biotechnology companies, academia, non-profit institutions, and international partners. The incorporation of technologies into the organ platforms, making them useful and attractive to a broad spectrum of the biomedical community and industry, is a goal of the MPS Program; adoption of the platform

technology for widespread use would be an important indicator of the program's success.

Future of drug screening

The MPS platforms are to be an attractive and viable option as a surrogate for human tissue in therapeutic testing and research. The integration of multiple organ modules to form a ''body-on-a-chip'' is the greatest advantage for this technology. A single organ chip will provide relevant results, but the integration of multiple organs will provide a wealth of additional results, including data related to issues such as organ–organ interactions, pharmacokinetics/pharmacodynamics, hormone/immune/cytokine responses to drug exposure, and physiological reactions to drug metabolites. This type of data obtained from medium to high throughput systems can accelerate drug discovery and screening for safe and effective compounds, reducing the number of costly and time-consuming clinical trials. The MPS organ system platforms can contribute to advancements in understanding the mechanisms underlying toxicity and the variability in toxic responses among individuals (i.e., demographic, genetic, epigenetic factors). Such insights would facilitate drug discovery by reducing the costs and time required to eliminate ''clinical failures'' and to focus on more promising potential therapeutics. Implementation of testing in human ''tissue-on-a-chip'' platforms could facilitate discovery of drug tolerance mechanisms, without the potential for adverse consequences associated with premature clinical trials. Such testing may be used to identify subpopulations that are tolerant to the use of drugs previously deemed ''unsafe'' on the basis of more generalized results (i.e., ''personalized medicine'').

Capitalizing on advances in iPSC technology

Linking organ modules together is not easy: compatibility, scaffolding material, flow rates, scaling and universal media must be considered for the integration of multiple organs. The MPS Investigators are addressing these issues to achieve the goal of creating a human-on-a-chip. As we plan for the future of the MPS Program, renewable cell sources originating from a single or a few primary sources, or generated (from genetically distinct or diseased donors) using standardized protocols will be necessary to assure the reliability and reproducibility of results. The use of induced pluripotent stem cells (iPSC) holds the greatest potential for addressing this requirement. Reprogramming easily obtainable tissue (e.g., skin fibroblasts or blood cells) into stem-cell-like sources is an optimal approach. The combined use of microphysiological systems on chips and iPSC technology opens the way to expanded, less-expensive drug screening and useful new tools for basic biomedical research that has proven difficult in the past. The combined MPS and iPSC approaches could significantly enhance individualized medicine, such as predicting responses to drugs or toxins, understanding genetic factors in disease, or the bases for autoimmune responses. The use of iPSC in microphysiological systems may prove useful in the treatment of patients with rare diseases, for which tissue sources are limited and large-scale clinical trials are impossible.

The use of iPSCs has much promise, but difficulties remain to be resolved. To date, no single iPSC line can be differentiated into all major organ tissues. In addition, various iPSC lines can differentiate into several cell types, but full maturation is extremely difficult. The need to improve iPSC technology for the development of reliable cell sources for the MPS Consortium is an integral part of the program; hence, a contract for a repository for the storage and distribution of well-characterized iPSC lines is being made available for use by the awardees. However, much work remains to develop such a resource for a human-on-a-chip. To ensure that iPSC cells are of high quality, MPS Program-generated resources have been allotted to characterize and standardize cell sources utilized in the platforms. Future ventures for iPSC development within the MPS Consortium will be to encourage further differentiation/maturation protocols, distribute cell lines across project teams and compare data obtained from such resource sharing that could ensure tissue quality in the microphysiological platform. Added benefits of such practices could also galvanize the overall iPSC field of study.

Additional uses for organs-on-chips

While the primary objective of the MPS Program is to create organs-on-chips for drug efficacy and toxicity screening, other areas of research could benefit from the use of the organ system platforms as additional research or clinical tools (Figure 1). For example, many infectious diseases require a human host for pathogenesis, making basic mechanistic studies difficult. If MPS platforms can replicate the human pathogenesis, they can serve as models for mechanistic studies, facilitating the development of effective treatments. Other potential applications include the identification or characterization of environmental toxins; studies using organ systems such as the gastrointestinal tract or bone marrow to determine the impact of biohazard and radiation exposure and to identify better countermeasures; well-controlled studies of the human microbiome; or fundamental studies of human regulatory physiology.

Future in scientific research

As the MPS Program advances, different project groups within the consortium will generate multiple versions of a human-on-a-chip. These may be iterative versions that incorporate improvements or they may be optimized for slightly different experimental or testing applications. The development, refinement and implementation of these platforms hold the potential for new discoveries with implications for human health, drug discovery and disease treatments. Implementation of humans-on-chips or organs-on-chips provides the basis not only for expedited efficacy and toxicity testing at a more rapid pace than previously envisioned, but also for reduced reliance on animal models for testing and studies of cellular mechanisms. Advances in platform technology will provide a pivotal research tool that is reliable, affordable and

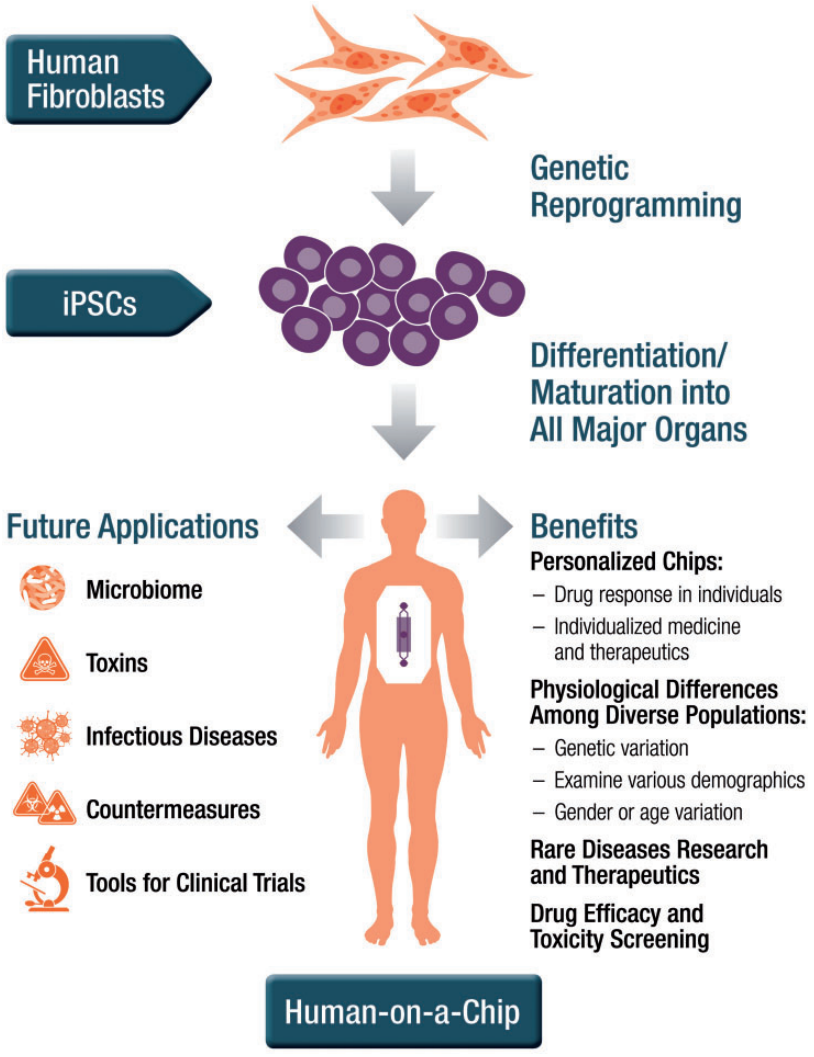

Figure 1 Potential for MPS technology. The use of iPSC lines will be an asset for organ module integration into a human-on-a-chip. Fibroblasts from an individual could be reprogrammed into iPSC lines, which can then be differentiated into renewable cell sources for all major organ tissues. Human-on-a-chip technology will allow a robust model for personalized drug responses, which could provide valuable insight into an individual's reaction to specific treatment regimens and compound tolerability. More in-depth knowledge could be gained with regard to various population differences, including genetics, gender and demographics. Even in special cases, such as patients with rare diseases, tissue samples could be obtained and examined to learn about mechanism and potential therapeutics. Future use: The MPS Program will produce organ modules as deliverables used for drug discovery and screening. However, the potential for MPS platforms could be utilized in many areas of research, including countermeasures, the microbiome, responses to toxins, infectious diseases and in conjunction with clinical trials

reproducible: ''Organs-on-chips'' derived from defined cell sources could provide basic scientists with more easily managed, less costly experimental models for studies that are not reliant on whole organism or behavioral responses. For example, studies easily adaptable to platform technology might focus on signaling pathways, the influence of epigenetic factors, or the responses of gut, neural or skin tissue to the microbiome. Translational science would be expedited by the use of the well-defined platforms for initial experiments to identify the molecular targets for drugs, or the basic mechanisms that influence drug metabolism or distribution within compartments such as the central nervous system. Clinical studies would be facilitated by the detailed pharmacokinetic data derived from humans-on-a-chip, and clinical treatment plans could be personalized on the basis of data derived from an ''individual-on-a-chip.'' The widespread potential utility of organs on platforms is to support more rapid advances in biomedical discovery and development of effective clinical treatments to improve human health.

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