IAC-19.A2.7.5x53999

Tissue Chips in Space

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Abstract

The International Space Station (ISS) U.S. National Laboratory in collaboration with the National Center for Advancing Translational Sciences (NCATS) and the National Institute of Biomedical Imaging and Bioengineering (NIBIB) at the National Institutes of Health (NIH) developed the "Tissue Chips in Space" initiative to promote and fund research into human physiology and disease in low Earth orbit (LEO) that will translate into advancements in Earth-based medicine. The Tissue Chips in Space initiative is part of NIH's larger Tissue Chip Program [1] that aims to develop bioengineered devices to improve the complex and expensive process of predicting whether drugs will be safe and effective or toxic in humans. These bioengineered devices, referred to as microphysiological systems, "tissue chips," or "organs-on-a-chip," leverage recent advances in cell biology, tissue engineering, and microfabrication to accurately model human organ tissues in *in vitro* platforms. These systems offer promising solutions for modelling human physiology and disease pathology for applications in areas where traditional cell culture and animal models fall short. This report provides an overview of the Tissue Chips in Space initiative, an update on the its current status, and a discussion of its potential long-term benefits.
Keywords: tissue chips, micro physiological systems, international space station, disease modeling, space flight effect

Acronyms/Abbreviations

Blood brain barrier (BBB)
Central nervous system (CNS)
Children's Hospital of Philadelphia (CHOP)
Defense Advanced Research Projects Agency (DARPA)
Induced pluripotent stem cells (iPSC)
International Space Station (ISS)
Massachusetts Institute of Technology (MIT)
National Center for Advancing Translational Sciences (NCATS)
National Institute of Biomedical Imaging and Bioengineering (NIBIB)
National Institutes of Health (NIH)
National Aeronautics and Space Administration (NASA)
Post-traumatic osteoarthritis (PTOA)
SpaceX (SpX)
University of California, San Francisco (UCSF)
University of Florida (UF)
University of Washington (UW)
U.S. Food and Drug Administration (FDA)

1. Introduction

1.1 The Collaborators

To expand the research opportunities available onboard the International Space Station (ISS), in 2005, Congress designated the U.S. portion of the ISS as a U.S. National Laboratory, facilitating access to space-based research and development for a broad range of commercial, academic, and government users. The ISS U.S. National Laboratory is responsible for managing all non-NASA research, and all ISS National Laboratory-sponsored investigations must have the capacity to utilize the ISS for Earth benefits. By engaging with other government agencies in the public sector and with universities and companies in the private sector, the ISS National Laboratory promotes and brokers a diverse range of research in life sciences, physical sciences, remote sensing, technology development, and education [2].

The National Center for Advancing Translational Sciences (NCATS) at the National Institutes of Health (NIH) was officially established in fiscal year 2012 to transform the translational science process so that new treatments and cures for disease could be delivered to patients faster. NCATS, one of 27 Institutes and Centers at NIH, strives to develop innovations to reduce, remove, or bypass costly and time-consuming bottlenecks in the translational research pipeline in an effort to speed the delivery of new drugs, diagnostics, and medical devices to patients [3].

The National Institute of Biomedical Imaging and Bioengineering (NIBIB) is authorized by the National Institute of Biomedical Imaging and Bioengineering Establishment Act H.R. 1795, which was signed into law on December 29, 2000. NIBIB's mission is to improve health by leading the development and accelerating the application of biomedical technologies. The institute is committed to integrating the physical and engineering sciences with the life sciences to advance basic research and medical care. [4].

1.2 The Problem

Human physiology and disease pathology involve complex interactions between different cells, extracellular matrices, cell signalling molecules, and other factors. These interactions occur both in specific microenvironments as well as on the larger systemic level. However, both healthy and disease modalities have traditionally been studied using reductionist approaches that include the use of two-dimensional cell cultures that are often void of their native extracellular environment and lack the three-dimensionality that cells maintain *in vivo*. In part, these reductionist approaches have been used to take advantage of advances in robotics and the adoption of high-throughput screening programs. While these simplified approaches are useful in some cases, such as for validating target proteins or pathways and identifying hit compounds for development, they risk providing results that are outside the context of the larger system that human diseases actually exist in.

On the whole systems side, non-human surrogates are often used to study disease pathology and validate therapeutic interventions. These surrogates include rodents, dogs, nonhuman primates, zebra fish, and larger mammals such as pigs. Although these animals provide many advantages for research, including the ability to model systemic responses in complex organisms, be subjects for genetic editing tools, and reduce the risk for first-in-human tests, animals have different physiological and genetic responses, and there is a push for development of novel human systems that faithfully model human physiology and reduce animal use in research [5-7].

Recent studies have shown that, on average, across all indications, there is less than a 14% chance that a drug entering clinical trials will succeed in receiving U.S. Food and Drug Administration (FDA) approval [8]. The primary source of drug failure is due to an inability to demonstrate efficacy [9]. Another main reason for failure is safety, which is also monitored post-approval and can result in the removal of a therapeutic from the market. While a number of factors can contribute to the failure of a clinical candidate, it is well-recognized that the lack of relevant and appropriate preclinical models is a factor in many cases.

2. Tissue Chips

With the recent advancements in cell biology, tissue engineering, and microfabrication, there has been a multidisciplinary approach to building systems that are more human-relevant for use in studying disease pathology and validating therapeutic efficacy and safety. As part of this effort, many groups have developed tissue chip platforms that utilize 3D human multicellular complexes. These complexes are engineered into a structural environment that attempts to replicate the native *in vivo* environment. Such tools provide a more useful model than traditional 2D cultures, and through the use of human cells (healthy, diseased, or engineered) can replicate patient-specific pathology and open up avenues for precision and personalized medicine approaches [10].

To date, numerous studies have utilized tissue chip platforms. Tissue chips have been used to recapitulate functional tissues from organs such as bone, brain, gut, liver, lung, skin, and others [11-15] and to model diseases [10], including rare diseases [16] such as Progeria [17] and Barth Syndrome [18]. Tissue chips have also been utilized to preform pharmacokinetic testing as well as efficacy and toxicity screening [19-21]. In addition, tissue chip platforms have been used to study cancer biology [22,23], and much progress has been made to link multiple tissue chips together to study interactions between different organs and model systemic responses [24-26].

The development of tissue chip technology has been supported by the NIH Tissue Chip Program, which issued 19 awards in 2012 to investigators to develop human tissue chips that would provide better predictions of drug safety and efficacy than current models. The program ran alongside a sister program supported by the Defense Advanced Research Projects Agency (DARPA) which focused on funding the development of 10 linked organ systems that would model system effects of disease and be useful for drug evaluation. The NIH Tissue Chip Program has now expanded to over 40 teams supported by more than 15 NIH Institutes or

Centers. The program is heavily collaborative with other government agencies such as the FDA and commercial partners from the pharmaceutical industry in order to expedite the translation of discoveries into therapeutics. In 2016, the ISS National Laboratory and NCATS entered into a collaboration to create the Tissue Chips in Space initiative, and in 2018, NIBIB joined the initiative [27].

3. Tissue Chips in Space

It is well documented that microgravity accelerates changes in human physiology, such as muscle wasting, osteoporosis, reduced cardiopulmonary function, and altered immune response, among others, and in many instances, these changes directly correlate to disease pathology on Earth [28-31]. By utilizing tissue chips containing human cells on the ISS National Laboratory, disease pathologies that might take years to produce on Earth are accelerated and can be studied on an expedited time frame.

To date, the Tissue Chips in Space initiative has funded nine projects, five of which were awarded from NIH RFA-TR-16-019 and four of which were awarded from NIH RFA-TR-18-001 (see Table 1). Additionally, as many of these changes are reversible upon return to Earth, these studies are suitable for developing drug targets. The awards, which are issued as cooperative agreements managed by NIH, require the teams to adhere to strict timelines and meet quantitative milestones in order to progress. The initiative provides the opportunity for each awarded team to launch two separate experiments to the ISS, contingent on successful completion of each project's milestones. The first phase of each project is designed to validate the disease model of study, and the second is to test novel therapeutics in the model. Each awarded team works with an Implementation Partner, a commercial company which is responsible for payload development and flight logistics such as manifesting and safety certifications.

As noted previously, the goal of the Tissue Chips in Space initiative is to use tissue chip platforms and the unique microgravity environment of the ISS to develop models of human disease, with the ultimate goal of expediting the discovery of therapeutics for people on Earth. In order to accomplish this goal, awarded teams leverage previous knowledge regarding the effects of spaceflight on the human body and how those effects translate to human diseases on Earth (see Table 1 for a summary of projects funded through the Tissue Chips in Space Program). For example, a team from the University of California, San Francisco (UCSF) is using the known dysregulation of the immune system and inflammatory responses associated with spaceflight as a surrogate model for Earth-based immunosenescence. The project is specifically investigating microgravity-induced aging of the immune system (generated through simulated microgravity and spaceflight) and its role in tissue-specific healing and regeneration.

A team of researchers from the Massachusetts Institute of Technology (MIT) is exploring putative therapeutics to treat post-traumatic osteoarthritis (PTOA). Utilizing a tissue chip system comprised of human cartilage, bone, and synovium that is challenged with inflammatory cytokines and an acute impact injury, the team will validate the system as an appropriate model for PTOA (see Fig. 1). Once validated, the team will use the model to test therapeutic options and monitor their effectiveness through the use of intracellular and extracellular biomarkers.

Fig. 1. Canadian Space Agency astronaut David Saint-Jacques works with the Cartilage-Bone-Synovium Tissue Chips in Space investigation. Image Credit: NASA

A University of Washington (UW) team is leveraging the ISS environment to study proximal tubule proteinuria and distal tubule kidney stone formation (see Fig. 2). Kidney dysfunction can result in serious health problems such as proteinuria, osteoporosis, and kidney stone formation in patients on Earth. However, disease progression is often slow and difficult to model *in vivo* in terrestrial studies. Using a human cell-derived kidney tissue chip system onboard the ISS, the team hopes to accelerate the onset of the disease states to produce a system that will allow for rapid translation to Earth-based therapeutics.

Fig. 2. NASA astronaut Anne McClain works inside the Life Sciences Glovebox with the Tissue Chips in Space Kidney Cells investigation. Image Credit: NASA

The blood brain barrier (BBB) is a critically vital component of the body that serves as a gatekeeper between the circulating blood in the brain and extracellular fluid in the central nervous system (CNS). A model that allows monitoring of the dysregulation of the BBB could provide applications for studying neurological disorders as well as the transport of drugs and toxins to the CNS. To this effect, a team of researchers from Emulate is developing an automated BBB tissue chip platform derived from human cells for use both on the ground and on the ISS (see Fig. 3).

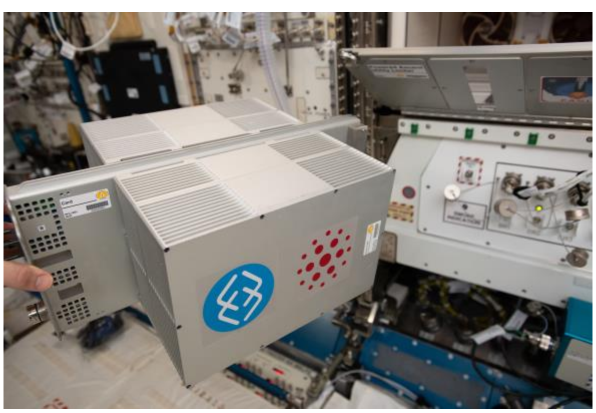

Fig**.** 3. The Organs-On-Chips as a Platform for Studying Effects of Microgravity on Human Physiology investigation is shown near the Powered Ascent Utility Locker. Image Credit: NASA

The innate immune response of humans allows for the recruitment of immune cells to an infected organ. A research team from the Children's Hospital of Philadelphia (CHOP) is developing a human airway tissue chip and connecting it to a bone marrow tissue chip. Using this interconnected system, the team can infect the airway chip and monitor the recruitment of neutrophils from the bone marrow as a model of innate response. Capitalizing on known spaceflight-induced changes associated with dysregulation of the immune system, this ISS project will serve as a model for a compromised immune system (see Fig. 4).

Fig. 4: The Lung Host Defence in Microgravity Chips in Space investigation hardware floats in the Destiny module of the International Space Station. Image Credit: NASA

Sarcopenia is characterized by the loss of skeletal muscle mass and function and commonly affects older adults. It is well documented that humans exposed to long-term spaceflight experience muscle wasting. A team from the University of Florida (UF) aims to use the accelerated muscle wasting environment of the ISS in combination with a human muscle tissue chip as a model for sarcopenia in terrestrial settings. Through the use of cells from different types of patients (human myocytes isolated from young, healthy and older, sedentary volunteers), the team will monitor the progression of the sarcopenia phenotype on a time scale not possible in terrestrial settings.

A team from Stanford University is utilizing human induced pluripotent stem cells (iPSC) from healthy patients to fabricate engineered heart tissue platforms for use on the ISS and on the ground. By taking advantage of microgravity-induced weakening of the heart muscle, the team will validate the ISS platform as a tool to model ischemic cardiomyopathy in humans on Earth. Once the model is validated, the team will use the platform to screen for potential drug candidates to treat patients. Similarly, in another project from the

University of Washington (UW), the team will use an engineered heart tissue chip to study aspects of cardiomyopathy associated with human health on Earth and in space. The project will utilize a novel magnetometer-based motion sensor to allow for automated real-time continuous functional readouts of the engineered heart tissues. The team will ultimately test pharmaceuticals and mechanical stimulations as potential therapeutic interventions.

Lastly, in a second project from Emulate, the team will use plug-and-play technologies to adapt their automated platform developed for their BBB ISS experiment to study dysregulation in the gut. The team will challenge the system with an infection and study the innate response and probiotic-induced response to the infection.

4. Conclusion

The work conducted through the Tissue Chips in Space initiative aims to leverage tissue chip technology onboard the ISS National Laboratory to lead to expedited discovery of therapeutics to treat patients on Earth. In addition, several ancillary benefits have already occurred as a result of the initiative, including the development of miniaturized automated systems for platform support. Sending tissue chips to the ISS requires the development of hardware that is automated and reliable with a small footprint. For example, in preparation for their spaceflight experiment, the University of Washington team, in collaboration with Implementation Partner Bioserve Space Technologies (see Table 1 for other implementation partners involved in the Tissue Chips in Space initiative), was able to reduce their tissue chip system from a 1,350-L volume system to a 45-L volume system [50]. In addition, tissue chip hardware for spaceflight must be designed to survive launch, operation onboard the ISS, and splashdown and must be capable of producing robust scientific results. The advancements that result from translating terrestrial systems to spaceflight systems are necessary steps in making tissue chip technology adaptable for wide-spread use in ground-based applications and to increase accessibility for greater numbers of researchers on Earth. One of the current limiting factors in the wide-spread adoption of tissue chip systems is the requirement for extensive hands-on interactions with the experiment. Through this program, this limitation is being addressed by development of automated and simplified systems such as these ones designed for spaceflight that allow for plug-and-play use. Overall, the need for these experiments to run in multiple terrestrial locations, as well as in orbit, without experimental confounders has pushed the biological and technical validation of the platforms, which translates into benefit for the whole field. As a result of the Tissue Chips in Space initiative, several commercial companies and other government agencies have begun projects that may utilize tissue chips on the ISS in order to address their own organizational mandates, further expanding and validating the use of tissue chip technology. Finally, while the primary purpose of the Tissue Chips in Space initiative is to translate results to benefit life on Earth, it is straightforward to imagine application of this technology to understand and mitigate the risks to human health posed by long-duration spaceflight.

Acknowledgements

We thank NASA for their support of this initiative. We also thank astronauts Anne McClain, David St-Jacques, Christina Hammock Koch, and Nick Hague for their laboratory support during Expedition 59. We also thank Amelia Smith (ISS National Laboratory) for editorial support.

Tables

Table 1. Summary of projects funded through the Tissue Chips in Space initiative. Principle Investigator (PI) refers to the PI listed as the Contact PI on the NIH grant. Awardee Organization is the institution awarded the management of the grant. UCSF = University of California at San Francisco, MIT = Massachusetts Institute of Technology, UW = University of Washington, CHOP = Children's Hospital of Philadelphia, SpX = SpaceX. Grant # is the current NIH Project Number associate with the grant (search https://projectreporter.nih.gov/reporter.cfm with the project number for additional details about each project). Spaceflight Effect is the known spaceflight effects the teams are looking to utilize in their models. Translation is the potential Earth-based therapeutic applications the projects are targeting. First Flight and Second Flight are the targeted launch vehicles for the given payloads. One project was launched on SpX-16 in 2018, and four were launched on SpX-17 in 2019. The remaining projects' launch vehicles and dates are currently to be determined (TBD).

Principle Investigator (Awardee Organization)	**Implementation Partner**	**Grant #**	**Spaceflight Effect**	**Translation**	**First Flight (year)**	**Second Flight (year)**
Sonja Schrepfer (UCSF)	Bioserve Space Technologies	UG3TR002192	Dysregulated immune system [32-34], inflammatory response [35,36]	Immunological Senescences [37,38], Adaptive Immunity [39], Impaired Myocardial Regeneration [40]	SpX-16 (2018)	TBD (2020)
Alan Grodzinsky (MIT)	Techshot	UG3TR002186	Musculoskeletal injuries and disease [41-44]	Post-traumatic osteoarthritis [45]	SpX-17 (2019)	TBD (2021)
Jonathan Himmelfarb (UW)	Bioserve Space Technologies	UG3TR002178	Dysregulation of kidney function [46-48]	Proteinuric chronic kidney disease [49,50]	SpX-17 (2019)	TBD (2021)
Christopher Hinojosa (Emulate)	Space Tango	UG3TR002188	Immune system deterioration [51], blood brain barrier integrity [52]	Multiple sclerosis [53], epilepsy [54]	SpX-17 (2019)	TBD (2021)
Scott Worthen (CHOP)	Space Tango	UG3TR002198	Dysregulated immune system [32-34], poor myeloid cell mobilization [55-56]	Treatment of airway infections [57]	SpX-17 (2019)	TBD (2021)
Siobhan Malany (UF)	Space Tango	UG3TR002598	Muscle atrophy [58-60]	Sarcopenia [61,62]	TBD (2020)	TBD (2022)
Joseph Wu (Stanford University)	Bioserve Space Technologies	UG3TR002588	Cardiomyopathy [63-65]	Cardiovascular disease [66,67]	TBD (2020)	TBD (2022)
Deok-Ho Kim (UW)	Bioserve Space Technologies	UG3TR028094	Cardiomyopathy [63-65,68]	Cardiac atrophy [69], cardiac arrhythmias [70]	TBD (2020)	TBD (2022)
Christopher Hinojosa (Emulate)	Space Tango	UG3TR002595	Gastrointestinal dysfunction [71,72]	Gut microbiome [73,74]	TBD (2020)	TBD (2022)

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