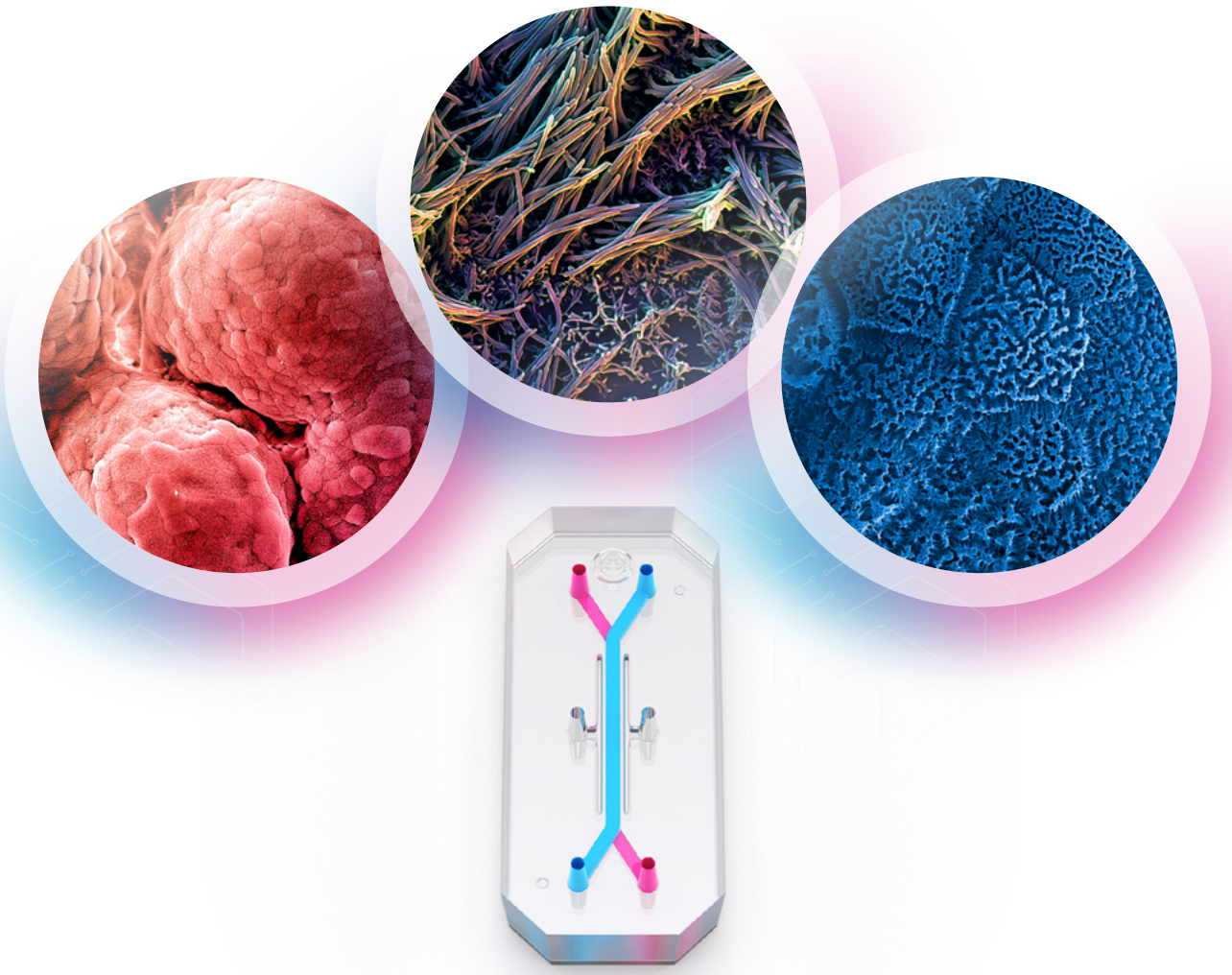

An Introduction to Organ-on-a-Chip Technology

emulate

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An Introduction to Organ-on-a-Chip Technology

Introduction

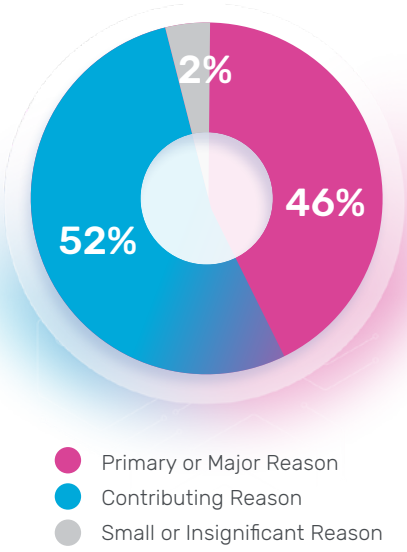
To bring drugs to market, scientists rely on different types of preclinical models—from 2D *in vitro* models to lab animals—that try to replicate human *in vivo* conditions. Unfortunately, conventional preclinical models have a difficult time approximating human biology and, as such, fall short in predicting how humans will respond to drugs. This is a major reason why 90% of drugs that pass the preclinical stage fail when they reach human trials.

Thankfully, scientists have next-generation technology at their disposal.

With Organ-on-a-Chip technology, researchers can test drugs in a human-relevant environment, get more accurate data on human response in a shorter time, and have greater confidence when sending drugs to clinical trials. Emulate has been leading the industry in bringing Organ-Chips to labs across the world since 2013. Over 100 peer-reviewed publications have demonstrated the utility of this technology in improving scientific understanding of human health and disease.

This guide is for researchers and scientists who are interested in learning how next-generation Organ-on-a-Chip technology can improve their disease research and drug development programs to increase efficiency and bring more life-saving treatments to market. After reading it, you'll have a better understanding of the most pervasive challenges in drug development as well as what Organ-Chips are, what they can do, and how they can help you overcome hurdles in developing new drugs.

98% of scientists say species differences contribute to high translational failure rates*



*2021 survey of 125 scientists conducted by the Linus Group.

Challenges with Preclinical Drug Development Models

To develop drugs, scientists rely on animals like mice, dogs, and non-human primates (NHPs) as well as *in vitro* models like 2D cell cultures and organoids/spheroids to predict human response. However, both *in vivo* and *in vitro* models come with unique sets of issues that can hinder the drug development process:

Challenges with Conventional *In Vivo* Models

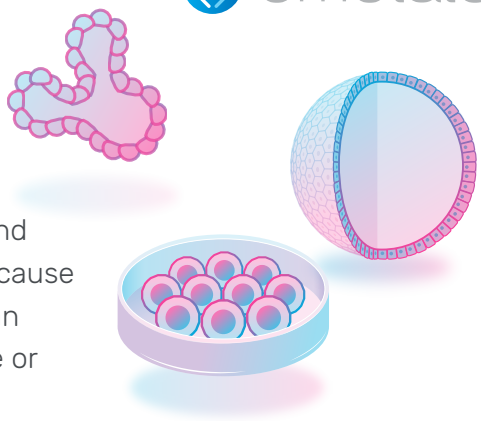
1. Difficulty in Sourcing NHPs: Since NHPs approximate human biology better than other lab animals, they are always in high demand; however, there is an ongoing shortage in NHP imports driving up demand and prices. There are many reasons for this, from extensive regulation, to a crackdown on breeding practices, to the lingering effects of the COVID-19 pandemic, but the shortage doesn't appear to be going away any time soon.
2. Lengthy and Rigid Animal Experiments: Experiments using animal models take a long time and require significant regulatory oversight. Each kind of animal model will bring its own logistical, regulatory, and ethical challenges that can further increase experiment costs and timelines.
3. Lack of Reproducibility: Regardless of the measures scientists take to standardize animal model experiments, each lab animal will be unique and highly susceptible to factors that scientists can't anticipate, which can impact study results. In fact, one Amgen study showed that only ~10% of *in vivo* experiments submitted as part of clinical development could be reproduced by an in-house group of researchers[1]. Even factors like the way scientists handle lab animals or the technician's gender can influence the way an animal will respond in experiments. This makes reproducibility, which is essential to any kind of research, difficult to maintain.
4. Species Translation Issues: Despite efforts made to humanize animal models, inherent differences in species biology will always be a limiting factor in predicting human response. In fact, 98% of scientists agree that species differences contribute to high translational failure rate[2]. As the drug pipeline shifts towards biologics, immune cell therapy, and other human-specific treatments, these challenges will only grow.

92% of Industry

95% of Academics

say their progress is limited **'sometimes'** or **more often** by conventional research models

2021 survey of 125 scientists conducted by the Linus Group

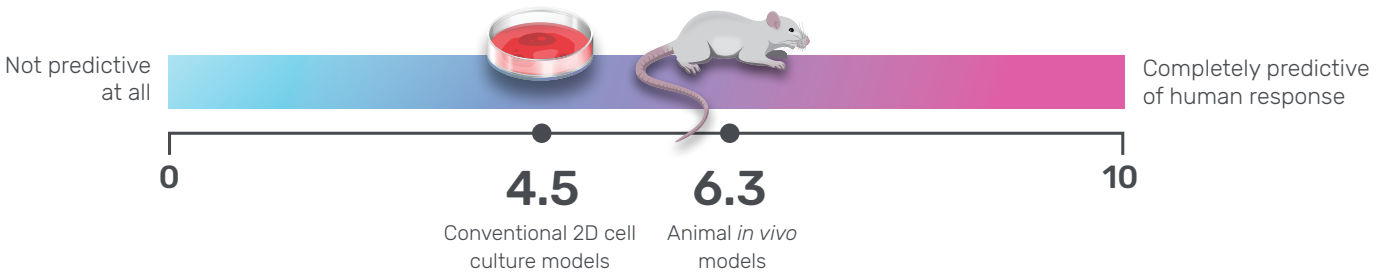


Challenges with Conventional *In Vitro* Models

1. Limited *In Vivo* Relevance of Immortalized Cell Lines: Scientists often rely on immortalized cell lines due to their ability to survive and continuously proliferate under cell culture conditions. However, because of the genetic manipulation needed to turn them immortal, they can behave differently than primary human cells, leading to incomplete or inaccurate understanding of disease biology and drug effects.
2. No Organ-Specific Microenvironment: *In vivo*, cells' functionality is heavily influenced by their environment—the extracellular matrix (ECM), mechanical forces, and stroma that surround them. Conventional *in vitro* models, however, lack these environmental features and can function differently than cells would *in vivo*.
3. Lack of *In Vivo* Complexity: In the human body, cells are constantly in communication with other cells. These interactions are integral to proper cell function. *In vitro* models, however, often only include a single cell type, leading them to respond to drugs in ways that do not reflect *in vivo* function.
4. Limited iPSC Differentiation: While induced pluripotent stem cells (iPSCs) hold significant promise for studying human biology outside of patients, it is difficult to unlock this potential. When cultured under static conditions, iPSCs often fail to differentiate fully into the cell type of interest.

Thankfully, many of these challenges can be overcome with next-generation Organ-on-a-Chip technology. Read on to learn more.

Scientists agree: Neither conventional cell culture models nor animal models adequately predict human response



2021 survey of 125 scientists conducted by the Linus Group

Organ-Chip Overview

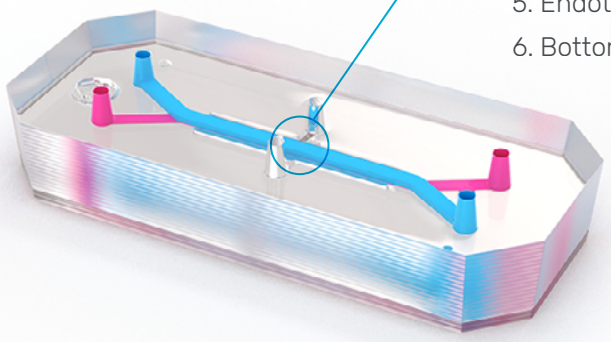
Organ-on-a-Chip technology allows researchers to recreate the functional unit of an organ using living human cells and an organ-specific microenvironment, offering a real-time window into the inner workings of human biology. The chips are small, flexible devices that contain two parallel channels. Many types of human-relevant cells can be seeded into these channels—including primary cells, iPSCs, organoids, and immune cells. The channels are separated by a thin, porous membrane, which creates a tissue-vascular interface for cell-cell communication. The membrane is coated with a tissue-specific extracellular matrix (ECM), helping to further drive tissue maturation as one would see *in vivo*.

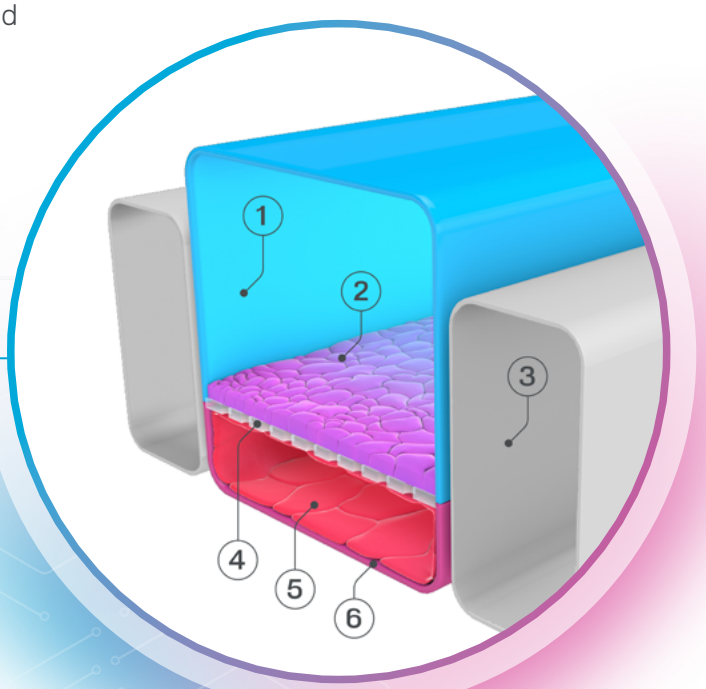
A key feature of Organ-Chips is that researchers can easily control and finely tune the mechanical forces cells experience. When Organ-Chips are placed under media flow and cyclic mechanical strain, cells experience the mechanical forces they would in the body—such as peristalsis in the intestines, breathing in lungs, and blood flow through vessels.

All of these features combined—multicellular complexity, cell-cell interactions, tissue-specific ECM, and mechanical forces—result in more *in vivo*-relevant gene expression, morphology, and functionality than is possible with conventional cell culture methods. Read on to learn where Organ-Chips fit into drug development workflows and how they can improve your odds of clinical success.

Schematic of an Organ-Chip

1. Top channel
2. Epithelial cells
3. Vacuum channel
4. Porous membrane
5. Endothelial cells
6. Bottom channel

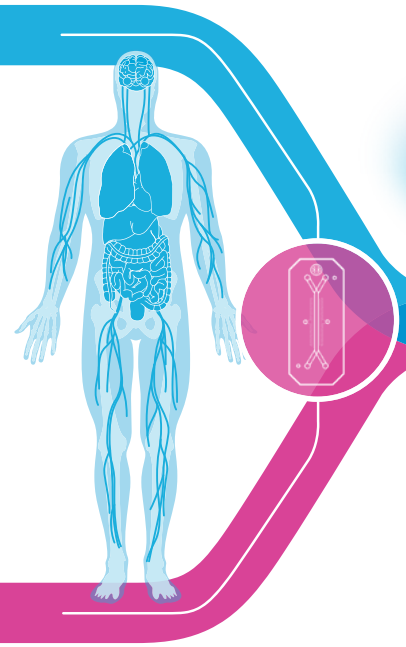


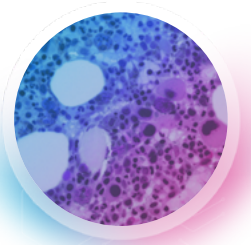


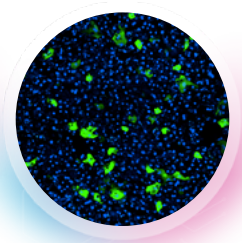
Organ-Chips consist of two parallel microfluidic channels (1, 6), with distinct channels for epithelial cells (2) and endothelial cells (5). A porous membrane (4) separates the two channels while enabling inter-channel communication and cell migration. Vacuum channels (3) alongside the microfluidic channels provide tunable mechanical stretch across the membrane.

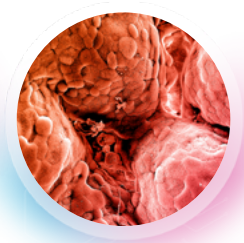
Organ-Chips in the Drug Development Workflow

With Organ-Chips delivering greater human relevance, the question becomes how to use them most effectively throughout the drug development pipeline. When we look at their applications, we see multiple benefits across the entire pipeline.



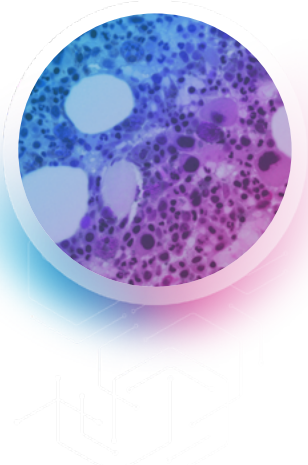






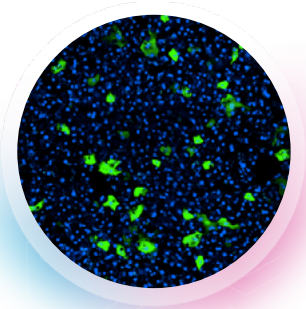
Early Discovery › **Lead Optimization** › **Preclinical Safety** › **Clinical Trials**

Early Discovery	Lead Optimization	Preclinical Safety	Clinical Trials
Develop disease models with closer-to-human gene expression to identify and validate drug targets.	Study candidate efficacy to rank order and optimize lead candidates.	Predict diverse mechanisms of unexpected human toxicity for preclinical drug candidates and assess human relevance of toxicity observed in preclinical animal studies.	Perform follow-up studies to assess efficacy mechanisms, or mechanisms of toxicity for drugs that have unexpected safety signals in the clinic.



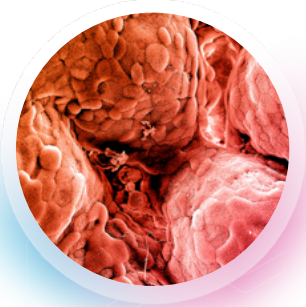
Early Discovery

In the early stages of drug discovery, Organ-Chips can be used to develop disease models with closer-to-human gene expression in order to identify and validate drug targets. In many experimental situations, two-dimensional cell culture methods are unable to replicate the appropriate cellular microenvironment, and animal models are unsuitable due to species differences. Organ-Chips have been used to generate human-relevant models for multiple organs and diseases, including bone cancer metastasis, bacterial vaginosis, thrombosis, environmental enteric dysfunction, and COVID-19, just to name a few. These models enable researchers to better understand underlying disease mechanisms and develop more effective treatments.



Lead Optimization

In the lead optimization phase, Organ-Chips provide human-relevant data to rank order and optimize lead candidates that show the most efficacy and least toxic profiles. By including Organ-Chip studies in this stage of drug development, a chemical compound that produced a toxic signal in a human Organ-Chip could be deprioritized from early *in vivo* studies, thus reducing animal testing and permitting safer candidates to progress through the development pipeline.



Preclinical Safety

In the preclinical safety phase, Organ-Chips can be used as a valuable screening tool to predict diverse mechanisms of unexpected human toxicity[3]. They can also be used in this phase to assess the human relevance of toxicity that may have been observed in preclinical animal studies[4]. Organ-Chips are particularly beneficial at this stage for evaluating newer drug modalities that are increasingly human-specific, such as biologics and immunotherapies.

Clinical Trials

Lastly, Organ-Chips can even benefit the clinical trial phase of drug development. For drug candidates that produce unexpected safety signals in the clinic, Organ-Chips can be used to perform follow-up studies in order to assess mechanisms of toxicity, and researchers can use that information to prioritize back-up compounds.

Organ-Chip Case Study

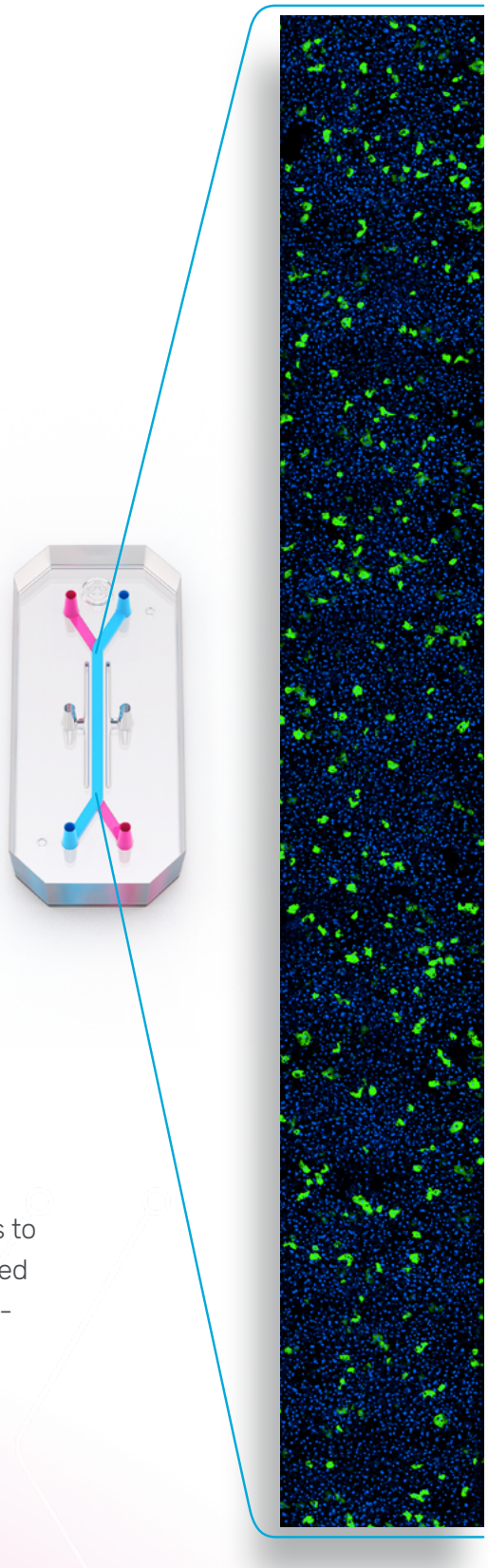
Researchers from Emulate worked with Janssen to evaluate the hepatoxicity of Atabecestat, a BACE inhibitor developed by Janssen that caused drug-induced liver injury (DILI) in clinical trials, by testing the compound on the human Liver-Chip. Through the study, they confirmed the hepatotoxicity of the compound, identified treatment-related mechanistic and injury biomarkers, and confirmed that the mechanism of action was oxidative stress triggering an inflammasome response. Ultimately, these results enabled Janssen to help screen safer back-up compounds.

As you can see, Organ-Chips can benefit the drug development pipeline at every stage. The next section of this eBook will further discuss the various insights that Organ-Chip experiments can provide.

Organ-Chip Insights

Each Organ-Chip enables researchers to collect and analyze rich, human-relevant data. Organ-Chips are compatible with endpoint assays used in conventional cell culture or in the clinic.

- Effluent-based analysis enables researchers to independently sample the effluent from each channel throughout an experiment. Effluent can be analyzed to measure relevant functional endpoints, such as biomarkers of tissue health and damage, apparent permeability (P_{app}), cytokine release, and metabolomics.
- Imaging analysis allows researchers to assess cell morphology, protein expression, and behavior, such as cell migration. Organ-Chips are compatible with a variety of imaging techniques, including brightfield, phase contrast, widefield fluorescence, confocal, multiphoton, and scanning electron microscopy. High-content imaging can be performed to capture the entire culture area across both chip channels, providing the maximum amount of data for quantification and downstream analysis.
- Omics-based analysis can be performed to assess Organ-Chip similarity to *in vitro* tissue or genetic differences between healthy and diseased tissue, enabling researchers to identify relevant disease pathways. Each chip contains enough genetic material for analysis on a chip-by-chip level, providing the opportunity for single-cell or bulk RNASeq as well as proteomic analysis.



In short, a wide variety of endpoints can be measured on Organ-Chips to better understand human physiology and disease. And through detailed endpoint protocols, Emulate can guide researchers throughout Organ-Chip analyses. Keep reading to learn more.

Applications

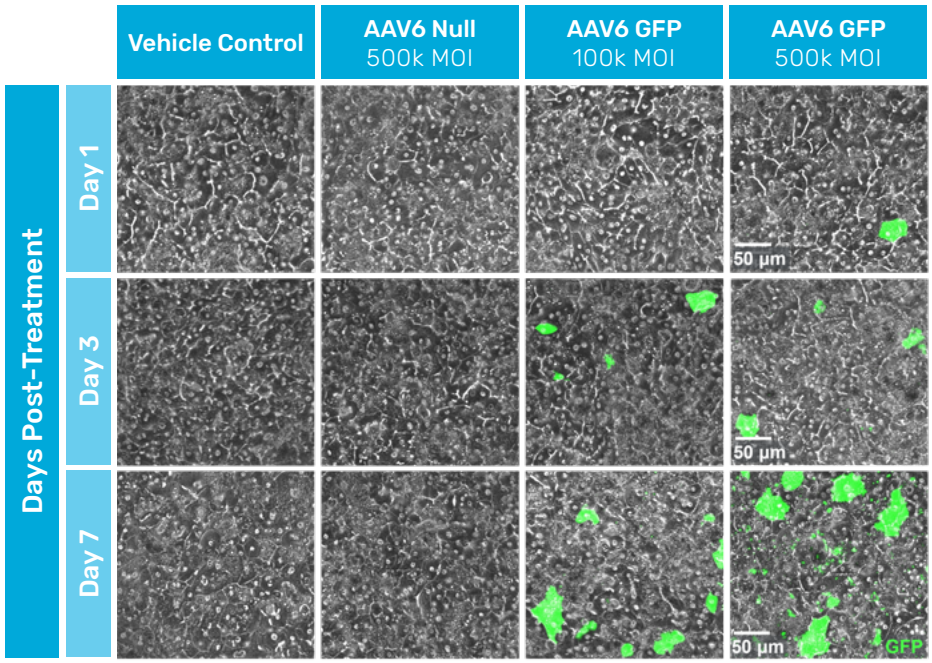
Here, we highlight a few of the many ways Organ-Chips are being used to provide more physiologically relevant insights into the mechanisms of human disease and the effects of drug candidates.

Gene Therapy

Gene therapy holds enormous promise for treating inherited and acquired genetic diseases, but progress in developing those therapies remains slow. Take for example adeno-associated virus (AAV)-based gene therapy: While there have been over 136 clinical trials for AAV-based therapies to date, there are only two approved therapies currently on the market. Additionally, 35% of those trials had severe adverse safety events, because conventional *in vitro* and *in vivo* models were unable to accurately predict toxicity.

With the AAV transduction application for the Emulate human Liver-Chip, researchers can rapidly iterate on AAV design in a human-relevant model of the liver sinusoid to accelerate vector optimization ahead of clinical trials. Users can administer a test AAV vector in the epithelial channel and monitor transduction efficiency and toxicity signals for up to seven days, enabling researchers to assess response in a time- and dose-dependent manner. Transduction efficiency can be quantified through GFP expression in the hepatocytes, while the potential toxicity of AAV vectors can be measured by functional biomarkers such as albumin secretion and ALT release levels.

AAV transduction is just one area within gene therapy where Organ-Chips can be applied. In addition to improving the development of AAV-based therapies, this workflow can be adapted to evaluate vectors other than AAV, including both viral and non-viral vectors. The flexibility and agility of this technology may be key to decreasing the timeline of bringing effective and safe gene therapies to patients. By enabling researchers to generate human-relevant data in just weeks—as opposed to the months it would take in non-human primates—Organ-Chips can facilitate rapid iterations on vector design to ensure that the most robust vector candidates proceed to clinical trials.



In hemophilia, AAV6 is one of the leading vectors for therapeutic research. To assess the use of the Emulate Liver-Chip for predicting AAV transduction and hepatoxicity risk, Liver-Chips were treated with AAV6 vector encoding green fluorescent protein (GFP). AAV6 demonstrated a time- and concentration-dependent transduction, while the hepatocytes and liver sinusoidal endothelial cells maintained a healthy morphology with no toxic signals present.

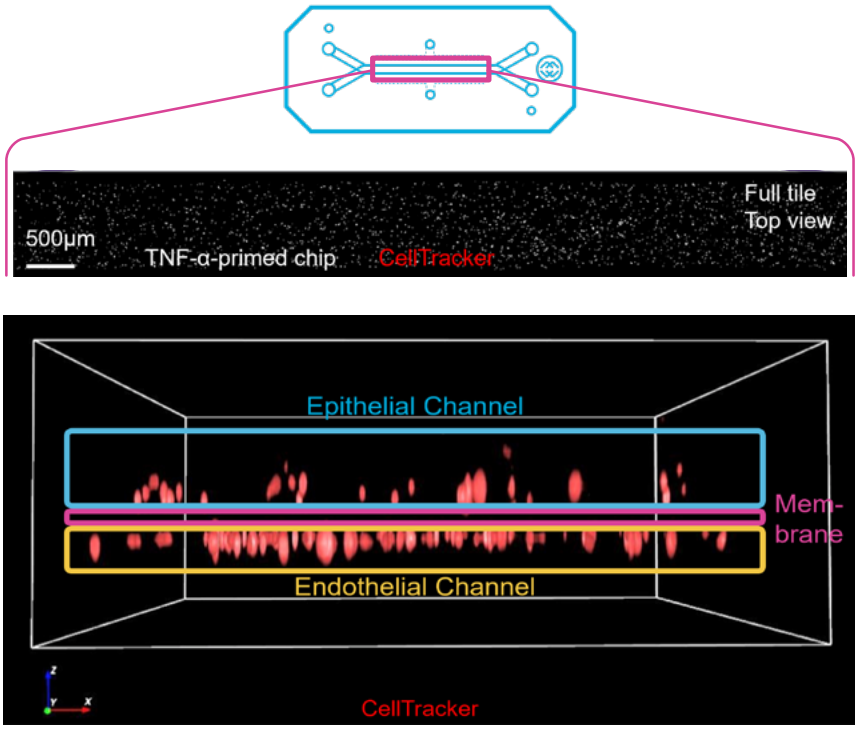
Learn more about testing the transduction of AAV-based therapeutics in the Liver-Chip.

Immunology & Inflammation

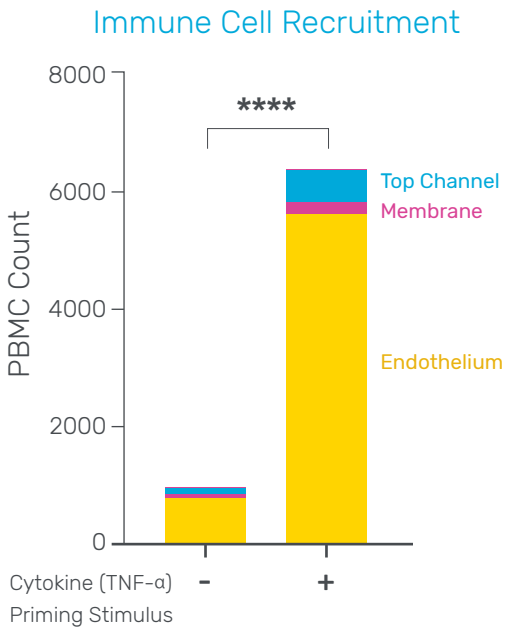
Inflammation plays a role in many prominent diseases, with chronic inflammatory diseases causing 3 out of 5 deaths worldwide[5]. However, the mechanisms of human inflammation remain poorly understood due to the challenge of modeling complex immune response *in vitro* and the fundamental differences in the immune systems of animals and humans. Because existing models can only capture individual aspects of immune response, researchers are left with an incomplete picture of the processes that drive human inflammation.

Organ-on-a-Chip technology can be used to build more human-relevant models of inflammation, allowing researchers to incorporate the cellular diversity seen *in vitro* into a tissue-specific and human microenvironment. The flexibility of Organ-Chip models allows researchers to precisely control and study the individual contributions of various factors in the inflammatory process—including resident and circulating immune cells, inflammatory cytokines, cell-cell interactions, and tissue-relevant mechanical forces—to better understand disease pathology and drug effects.

Take inflammatory bowel disease (IBD), for example. The Emulate human Colon Intestine-Chip can accurately model dysregulated immune cell recruitment, the driver of IBD pathogenesis. When immune cells flow through the chip vascular channel in the presence of proinflammatory stimuli, they undergo the entire process of immune cell recruitment—from attachment, to migration, to downstream effector function and barrier damage. This is the only model demonstrated to capture this full process in a tissue- and inflammation-specific manner, enabling a more complete window into disease pathogenesis. Four clinically relevant IBD therapeutics with different mechanisms of action have been assessed on the model, showing that Organ-Chips can be used to evaluate drug efficacy and rank order lead candidates.



One-way ANOVA of full dataset,
n=5, ****p ≤ 0.0001



Complex immune response, including immune cell recruitment, can be modeled. Shown here is the recruitment of PBMC immune cells from the vascular channel into the epithelial channel—a key step in IBD pathogenesis.

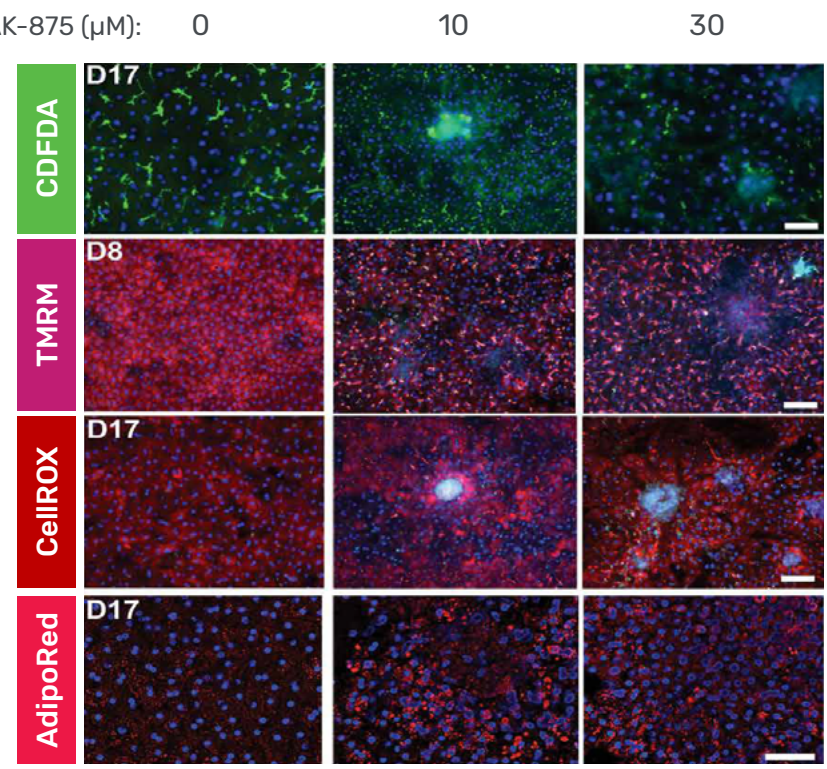
Learn more about modeling inflammatory immune cell recruitment.

Toxicology

Biopharmaceutical companies encounter many challenges in developing safe and effective drugs, but evaluating human toxicity can be particularly challenging. Approximately 30% of drugs fail during clinical trials due to toxicity—despite having passed preclinical safety screenings in animals[6]. Put simply, conventional models lack the predictive value required to confidently transition drug candidates to the clinic.

Organ-Chips enable researchers to translate to the clinic with confidence by predicting human response earlier in drug development. To measure how much Organ-Chips could improve patient safety, researchers qualified the Emulate human Liver-Chip against the guidelines defined by IQ MPS, an affiliate of the International Consortium for Innovation and Quality in Pharmaceutical Development. The study demonstrated that the Liver-Chip was able to correctly identify 87% of the tested drugs that caused drug-induced liver injury (DILI) in patients despite passing animal testing evaluations. At the same time, the Emulate human Liver-Chip did not falsely identify any drugs as toxic, supporting its use in toxicology screening workflows[3].

To demonstrate how using Organ-Chips in preclinical workflows can improve outcomes in clinical trials, consider this use case: TAK-875 was a drug candidate discontinued during phase III trials due to DILI. When the compound was retrospectively studied on the Liver-Chip, results showed that prolonged exposure to the compound caused mitochondrial dysfunction, oxidative stress, lipid droplet formation, and an innate immune response—all harbingers of DILI for susceptible patients[7]. Had the Liver-Chip study been performed prior to clinical trials, researchers could have found human-specific toxicity concerns earlier, deprioritized TAK-875 as a drug candidate, and moved forward with safer candidates.



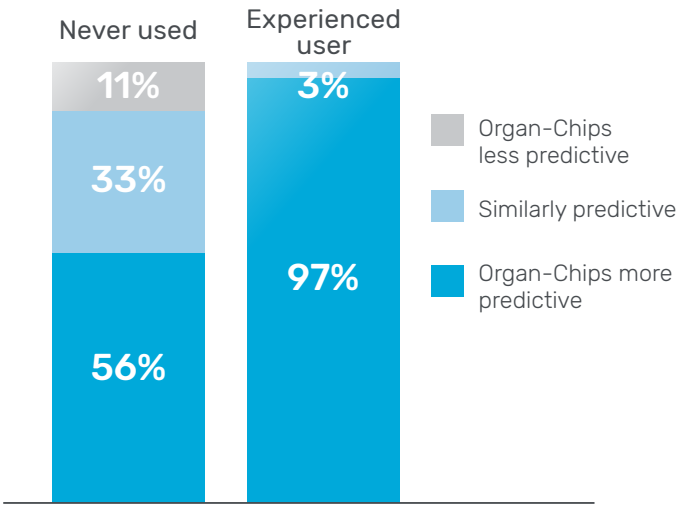
Identifying risk for idiosyncratic DILI. Human Liver-Chips were treated daily with TAK-875 at the equivalent of human C_{max} (10 μM) to determine if they could detect DILI. Confocal microscopy analysis showed that treatment resulted in a dose-dependent decrease in MRP2 activity, as measured by biliary efflux of the MRP2 substrate CDFDA (green). It also showed that treatment had an effect on mitochondrial membrane potential, confirmed by a dose-related and time-dependent redistribution of tetramethylrhodamine methyl ester (TMRM), lipid droplet accumulation, and formation of reactive oxygen species (ROS). These results align with the clinical outcome of TAK-875 treatment, supporting the use of detecting idiosyncratic DILI in the Liver-Chip.

Learn more about assessing compound toxicity with Organ-Chips.

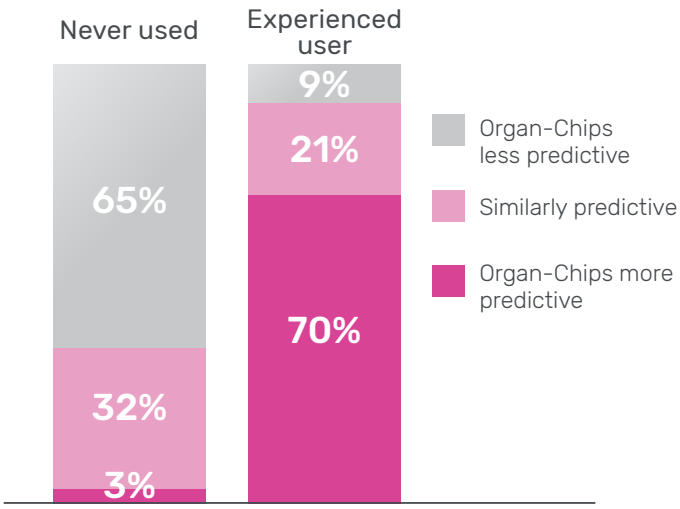
Organ-Chip User Survey Results: A Comparison to Conventional Preclinical Models

Organ-Chips are such a powerful technology because they can help scientists predict human response earlier throughout drug discovery and development. Experienced Organ-Chip users agree, with 97% saying Organ-Chips are more predictive than conventional *in vitro* models. The more surprising finding, however, is that 70% of experienced Organ-Chip users rate the technology as more predictive than *in vivo* models, with an additional 21% rating the technology as similarly predictive[2]. These survey results speak directly to the impact that incorporating Organ-on-a-Chip technology can have on one's research program.

Opinions on Organ-Chips vs Conventional Models



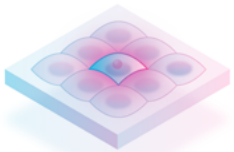
Organ-Chips vs Conventional *In Vitro* Models

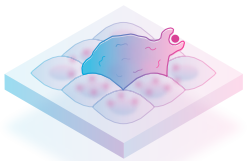


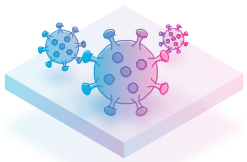
Organ-Chips vs *In Vivo* Models

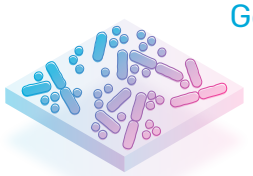
Explore All Emulate Organ-Chip Applications by Clicking Below


Cancer

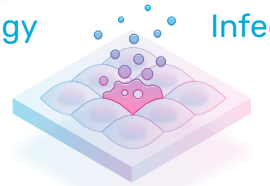

Gene Therapy


Immunology


Infectious Disease


Microbiome


Neuroscience


Toxicology

Key Learnings & Additional Resources

As discussed throughout this eBook, Organ-on-a-Chip technology can help researchers overcome several challenges associated with conventional *in vitro* and *in vivo* models. The following table summarizes some of the most common challenges and how Organ-Chips can help.

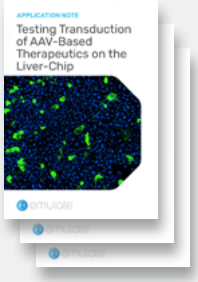
Model Type	Challenge	How Organ-Chips Can Help
Conventional *in vivo* models	Difficulty in sourcing NHPs	Organ-Chips provide human-relevant insights and can minimize the number of NHPs needed by enabling researchers to perform lead optimization studies in Organ-Chips before proceeding to NHP studies.
	Lengthy and rigid experiments	Organ-Chip studies can be designed and executed without any regulatory oversight required. Additionally, Organ-Chip studies can be easily adjusted to adapt to insights gained throughout the study.
	Lack of reproducibility	Organ-Chip kits with pre-qualified human cells enable researchers to generate robust, reproducible results.
	Species translation issues	Organ-Chips use physiologically relevant human cells to avoid species translation challenges.
Conventional *in vitro* models	Limited *in vivo* relevance of immortalized cell lines	Organ-Chips are compatible with a wide variety of human-relevant cell sources, including primary cells, iPSCs, and organoids.
	No organ-specific microenvironment	Organ-Chips recreate organ-specific microenvironments by incorporating a tissue-specific ECM, microvasculature, and the relevant biomechanical forces caused by fluid flow and stretch.
	Lack of *in vivo* complexity	Organ-Chips can incorporate multiple cell types, including epithelial cells, endothelial cells, resident and circulating immune cells, and even microbes to recapitulate the complex cellular interactions that occur *in vivo*.
	Limited iPSC differentiation	Organ-Chips improve iPSC differentiation by providing a more physiologically relevant microenvironment to drive gene expression that more closely resembles organ-specific *in vitro* transcriptomes.

To make Organ-Chips accessible and user friendly for researchers, Emulate has developed the lab-ready Human Emulation System® consisting of instruments, consumables, and software. With this complete solution, researchers can use Organ-Chips to replicate the biology and function of any organ.

Continue your Organ-Chip journey by exploring new topics at the following links:

Learn more about the Human Emulation System

See how to get started with Organ-Chips



Read examples of how Organ-Chips have been used

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