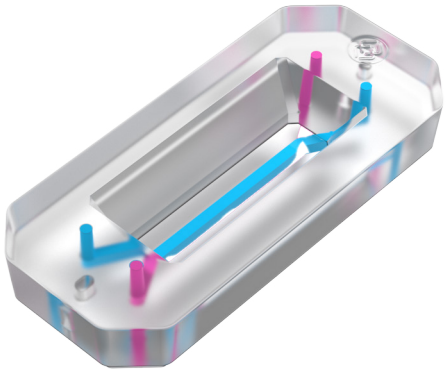
Chip-R1™ Basic Research Kit

Improve precision in ADME and Toxicology applications with a low drug-absorption profile



Overview

An Organ-Chip is a living, micro-engineered cell culture system that recreates the natural physiology and mechanical forces that cells experience within the human body. The Chip-R1™ Rigid Chip was designed to minimize drug absorption and enhance biological modeling by using low-drug-absorbing materials and featuring increased shear stress in the vascular channel. Constructed using rigid plastics, including a polycarbonate tissue culture membrane, Chip-R1 builds upon the foundation of microfluidic Organ-Chips, offering researchers greater precision in predicting human drug responses. With the Emulate Chip-R1 Basic Research Kit, users can build a wide variety of non-stretch organ models for ADME and toxicology applications without the concern of compound absorption by the microfluidic system.

Kit Components

The Chip-R1 Basic Research Kit contains all the non-biological components required to create an Organ-Chip for the Zoë Culture Module. Each kit consists of Chip-R1 Rigid Chip consumables, chip carriers, and Pod® Portable Modules (**Figure 1**). Chip carriers securely connect the chips to the Pods, while the Pod reservoir lid acts as the interface between Zoë and the chip.

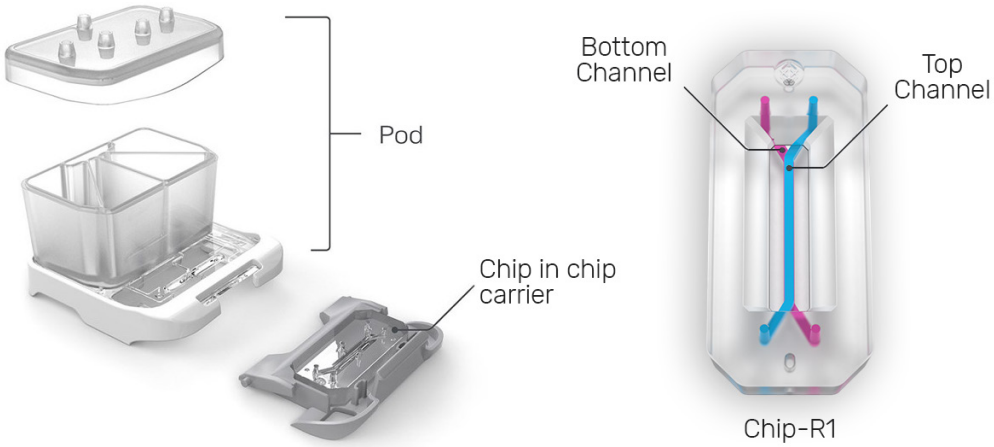


Figure 1: *Chip-R1 Basic Research Kit Components.*

Chip-R1 Configuration

The Chip-R1 Rigid Chip (**Figure 2**) is made of rigid plastics that minimize compound absorption. Chip-R1 retains much of the same microfluidic architecture of the Chip-S1® Stretchable Chip, including co-culture to model the tissue-tissue interface, independent perfusion of the apical and basal channels, and compatibility with Emulate's instrumentation hardware. With a polycarbonate tissue culture membrane, Chip-R1 is better suited for organ models that do not require mechanical stretch to mimic physiologically relevant strains cells experience *in vivo*—like peristalsis in the intestine or inflation of the lungs—such as liver and kidney.

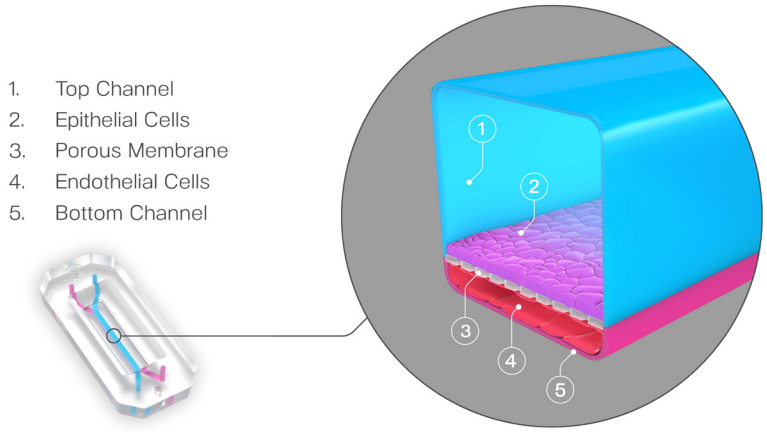


Figure 2: *Chip-R1 diagram.*

Pod Portable Module

Each Chip-R1 connects to a Pod-2™ which, through Zoë, enables automated control of media flow and dosing while maintaining ease of portability for routine microscopy observation (see **Figure 3**). The Pod stores 4 mL of media for each microfluidic channel, enabling automated media flow for up to five days. Media effluent can be easily collected from the outlet reservoirs for downstream analysis.

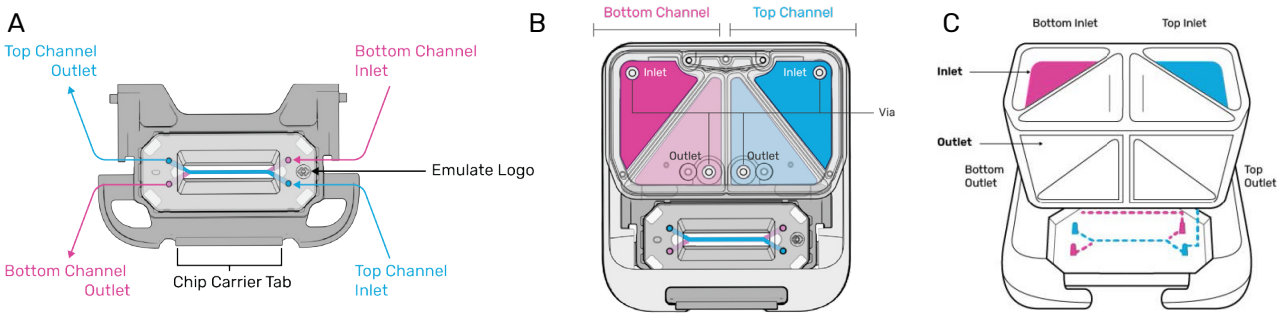


Figure 3: A) *Chip carrier diagram showing direction of fluid flow through the chip.* ***B)*** *Pod diagram showing reservoirs for top and bottom channel inlets and outlets.* ***C)*** *Diagram demonstrating fluid flow between Pod and chip.*

Drug Absorption Characterization of Chip-R1

Many microphysiological systems (MPSs), including Organ-Chips, are made with polydimethylsiloxane (PDMS) due to its beneficial properties such as transparency, flexibility, biocompatibility, and oxygen permeability. However, this material is known to absorb a subset of small hydrophobic molecules, which can interfere with the assessment of drug efficacy, toxicity, and ADME. Chip-R1 overcomes these limitations by using non-drug-absorbing rigid plastics, ensuring greater accuracy for evaluating drug candidates while building on key microfluidic capabilities.

To characterize the improvement in mitigating compound absorption, a panel of eight compounds was tested on both Chip-R1 and Chip-S1 chips. These compounds were selected for their relevance to hepatotoxicity or liver metabolism functions as well as for their range of physiochemical properties (**Table 1**), including hydrophobicity (logP), molecular weight, and topological surface area.

Compound	Dosing concentration	logP	Molecular Weight (Da)	Topological Polar Surface Area (Å2)	pKa	Ionization status at pH 7.4
Nefazodone	30 µM	4.3 - 4.7	470	51.6	7.09	Neutral
Diclofenac sodium	20 µM	4.26 - 4.75	318.1	52.2	4	Anionic
Bufuralol HCl	10 µM	2.99 - 3.24	297.82	45.4	13.06	Cationic
Midazolam	20 µM	2.73	325.8	30.2	5.5	Neutral
Labetalol	137 µM	2.7 - 3.1	328.4	95.58	9.3	Cationic
S-mephenyt-oin	35 µM	1.64 - 1.69	218.3	49.4	8.1	Neutral
Phenacetin	30 µM	1.41 - 1.62	179.2	38.3	14.98	Neutral
Trovafloxacin	200 µM	0.3 - 0.9	416.4	99.8	5.9	Zwitterion

Table 1: *Compounds assessed for absorption on Chip-S1 and Chip-R1.*

These compounds were applied to either the top or bottom channels of acellular Chip-S1 and Chip-R1 consumables at a flow rate of 30 µL/h. To determine the average compound recovery after 24 hours, effluent was collected from Pod reservoirs (inlets and outlets) for the 24-28 hour exposure interval, since a four-hour time interval was required to provide sufficient volume for LC/MS analysis. Compound recovery is reported as the average recovery for the four-hour time interval and represented as a ratio of Pod outlet concentration to Pod inlet concentration (**Figure 4**).

Drug Absorption Characterization of Chip-R1 (Continued)

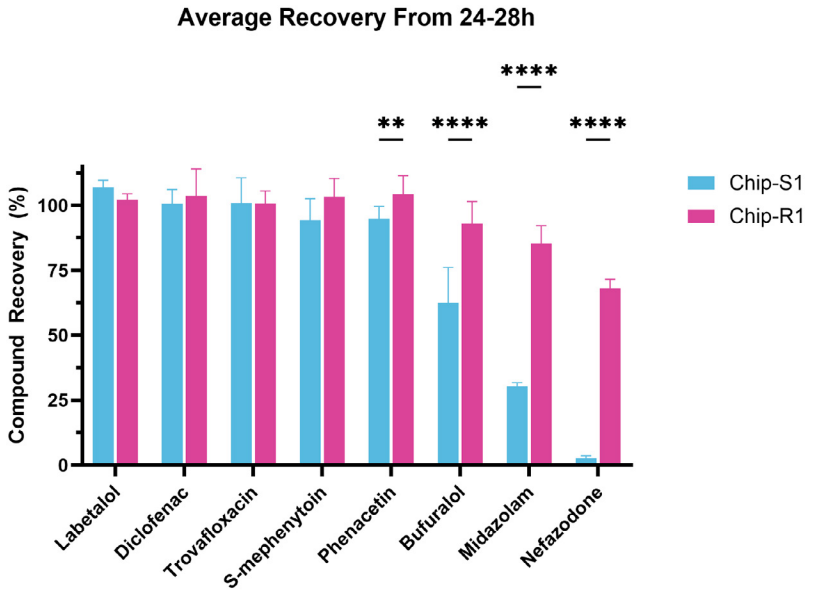


Figure 4: **Average Compound Recovery Between 24–28 Hours of Flow.** *Data shown as mean ±. Significance in recovery determined by two-way ANOVA with Fisher's LSD multiple comparison test. n=3–4 chips per condition, with values for top and bottom channels average per chip, **p<0.01, p<0.0001. assessed for absorption on Chip-S1 and Chip-R1.*

Out of the eight compounds tested, three demonstrated high absorption on Chip-S1: Nefazodone, Bufuralol, and Midazolam. These compounds would be predicted to have PDMS absorption liability based on their relatively high hydrophobicity, low molecular weight, and low topological polar surface area. Chip-R1 demonstrated a significantly improved recovery profile of PDMS-liable compounds after 24 hours versus Chip-S1:

- Bufuralol improved from 60% to 93% recovery
- Midazolam improved from 25% to 85% recovery
- Nefazodone improved from 2.7% to 68% recovery

The remaining compounds tested demonstrated no significant absorption on either chip.

Although Nefazodone showed an impressive improvement—with a 26-fold increase in recovery on Chip-R1 compared to Chip-S1—it is important to understand the absorption kinetics on Chip-R1 for compounds that still show some absorption liability. Thus, with Nefazodone being the most prone to absorption from the set of compounds tested, a more in-depth time course analysis was performed to better understand its absorption behavior in Chip-R1. As shown in **Figure 5**, Chip-S1 demonstrated negligible recovery of Nefazodone over the course of 48 hours, indicating that the PDMS-based chip absorbed most of the compound for the duration of treatment. While Chip-R1 demonstrated low recovery rates within the first few hours of dosing, after 48 hours, the rate of recovery was greater than 80% on average across both top and bottom channels. Extrapolated out to 72 hours, one would expect to see >90% recovery of Nefazodone. Together, these datasets demonstrate Chip-R1's utility in ADME and toxicology applications, even for compounds that are highly prone to PDMS absorption such as Nefazodone.

Drug Absorption Characterization of Chip-R1 (Continued)

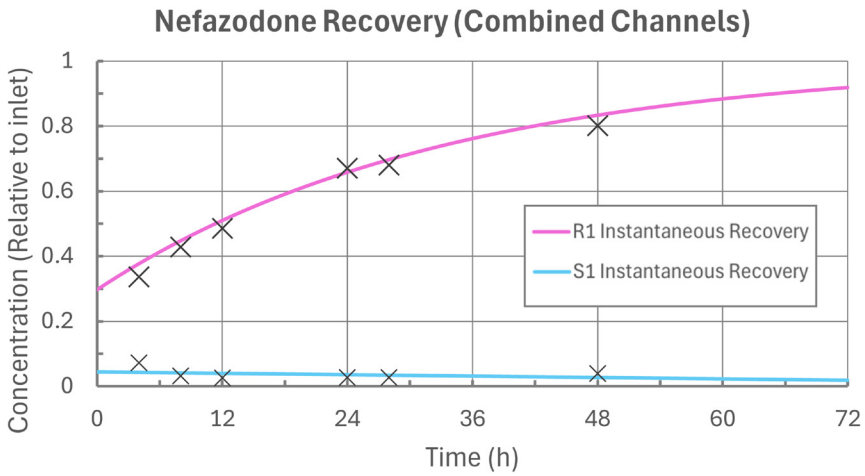


Figure 5: Absorption kinetics of Nefazodone on Chip-R1 versus Chip-S1. *Compound recovery was calculated by taking the ratio of outlet to inlet concentration. Marked X's denote measured data averaged over a four-hour time interval. Solid lines represent curve fit for instantaneous recovery values. n=3-4 chips per condition.*

Additional Features and Benefits

More Physiologically Representative Modeling

The unique features of Chip-R1 can improve research in areas beyond ADME and toxicology. Chip-R1 features a shorter bottom channel height at only 100 µm tall, which in conjunction with a maximum flow rate of 2,000 µL/h, enables researchers to achieve a maximum shear stress of 2.3 dyn/cm^2. The modified design of the vascular channel enables physiologically relevant levels of shear stress to be applied, which is critical for applications such as immune cell recruitment. Additionally, Chip-R1 features a thinner membrane at just 22 µm thick with 3 µm pores, enabling enhanced cellular crosstalk and better approximation of the *in vivo* milieu.

Shorter Imaging Distance to the Top Channel

Chip-R1 features a distance of only 172 µm from the bottom of the chip to the top of the tissue culture membrane, thus requiring a significantly shorter working distance to reach the epithelial layer (**Figure 6**).

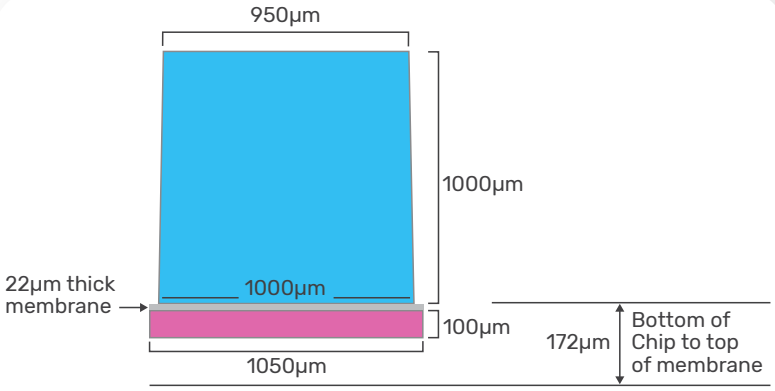


Figure 6: *Cross-sectional diagram of Chip-R1 illustrating imaging distance required to reach the top channel.*

Compatible with Zoë Culture Module®

The Emulate Chip-R1 Basic Research Kit is designed to be used with the Zoë Culture Module, a complete Organ-on-a-Chip platform that provides the dynamic conditions needed to culture up to 12 Organ-Chips per Zoë.

In addition to the Chip-R1 Basic Research Kit, Emulate offers additional companion products:

- Pod Imaging Adapter: for microscopic inspection during an experiment
- Fixed Chip Imaging Adapter: for post-experiment fixation, staining, and analysis

Chip Specifications:

Top Channel

Width x height dimensions	1,000 µm x 1,000 µm
Area at membrane	26.66 mm^2
Channel volume	24.52 µL
Imaging distance from bottom of chip to top of membrane	172 µm

Bottom Channel

Width x height dimensions	1,050 µm x 100 µm
Area at membrane	29.46 mm^2
Volume	2.97 µL

Membrane

Pore diameter	3.0 µm
Pore spacing	Random distribution (Track-etched membrane)
Porosity	2.8%
Thickness	22 µm

Kit Specifications:

Specification	Details
Compatible cell types	The open nature of the Emulate Chip-R1 Basic Research Kit makes it compatible with practically any type of human or animal cells, including: • Primary cells • Dissociated organoids • Induced pluripotent stem cells (iPSCs) • Immortalized cell lines Due to the rigid nature of the consumable, Chip-R1 is not recommended for organ models that require stretch.
Characterization endpoints	• Image analysis o Brightfield and fluorescence microscopy • Omics analysis o RNAseq, proteomics, and metabolomics • Effluent analysis o Cytokine release, injury markers, barrier function (P_{app}), etc.
Storage conditions	Ambient temperature (15–25°C)
Shelf life	1 year from date of manufacture
Sterility	All consumables—including chips, Pods, and carriers—are sterilized prior to shipment.

Zoë Culture Module Firmware Requirements:

Using Chip-R1 with your Zoë Culture Module may require upgrading your Zoë's firmware. Please refer to the chart below to determine whether your system will require a firmware upgrade prior to running Chip-R1 experiments.

Zoë Device	Chip R1 Version Info	Upgrade Path
Zoë-CM1®	Display Firmware: 1.3.2 Core Firmware: 1.2.5	**Any Zoë-CM1** user can upgrade their Zoë display and core firmware using Utility Hub 1.0 software from the Emulate website.
Zoë-CM2®	Display Firmware: 1.3.2 Core Firmware: 1.4.5	**Firmware 1.3.2:** Any user with a Zoë-CM2 running this version of firmware can self-upgrade using Utility Hub 1.0 software from the Emulate website. ***Below* firmware 1.3.2:** Any user with a Zoë-CM2 with a version of firmware below 1.3.2 cannot self-update. Please contact Emulate Support (support@emulatebio.com) for further assistance.

Ordering Information

Kits are available in sets of 12 or 24 chips. Each set contains:

- 1 Chip-R1 Rigid Chip in a chip carrier
- 1 Pod Portable Module

Product Name	Kit Contents	Catalog Number
Chip-R1 Starter Pack (6-pack)	6 sets of chips, 2 Chip Cradles, 10 square Petri dishes, plus 4 Steriflip® filters	STP-R1-6
Chip-R1 Basic Research Kit (12-pack)	12 sets of chips, plus 6 Steriflip® filters	BRK-R1-12
Chip-R1 Basic Research Kit (24-pack)	24 sets of chips, plus 8 Steriflip® filters	BRK-R1-24

Companion Products:

Product Name	Description	Catalog Number
Pod Imaging Adapter	Organizes 2 Pods for quick cell viability and health checks and ensures compatibility with SBS footprint inverted microscopes	POD-IMG
Fixed Chip Imaging Adapter	Organizes 12 fixed chips for high-throughput imaging and ensures compatibility with SBS footprint imaging equipment	CHIP-IMG