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Challenge of Standardization in the AMED-MPS Project

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Global Summit of Regulatory Science 2020

COI Disclosure

The authors have no conflict of interest to disclose with respect to this presentation.

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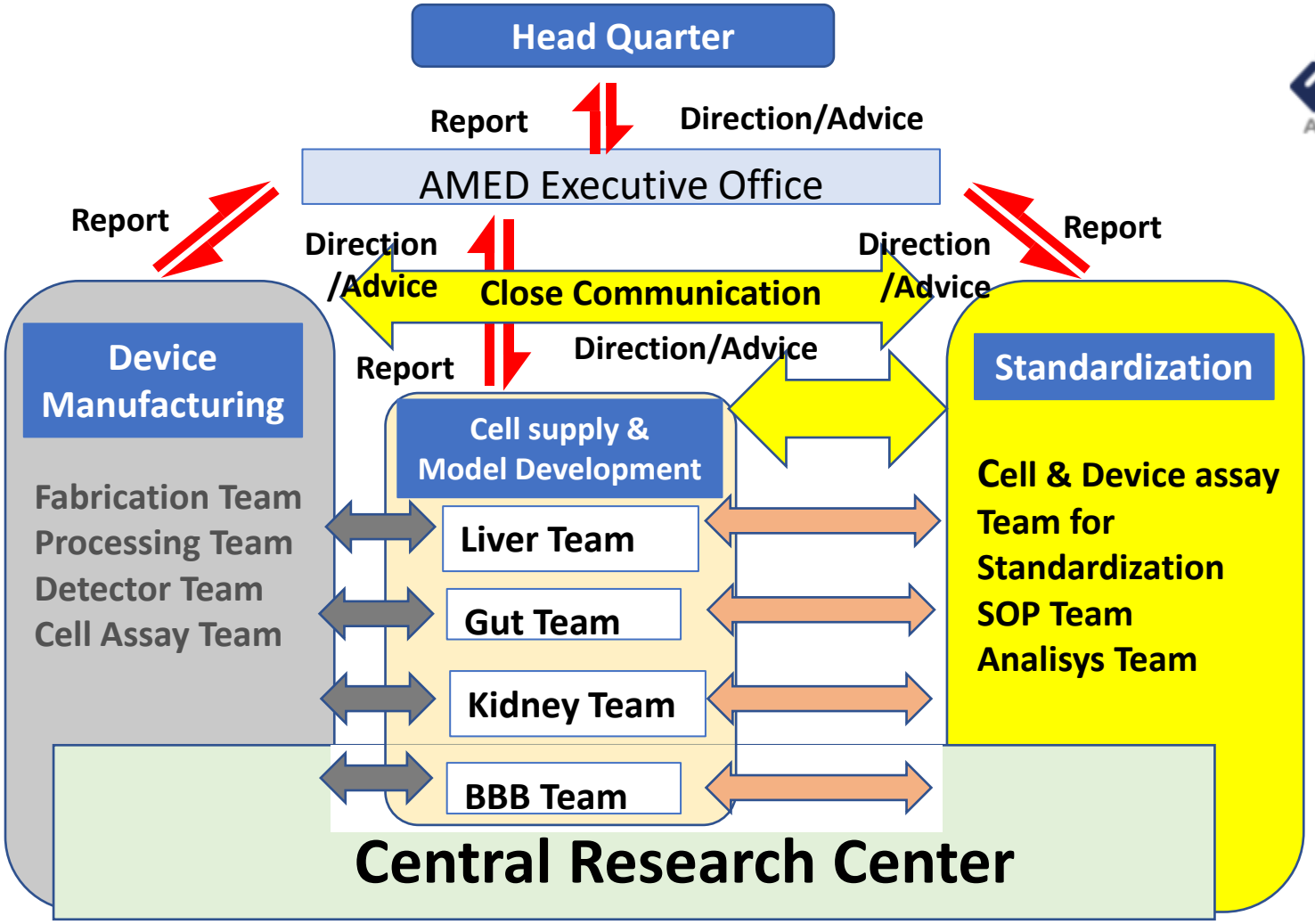
Seiichi Ishida

Microphysiological system (MPS) Project in Japan



Today's Agenda

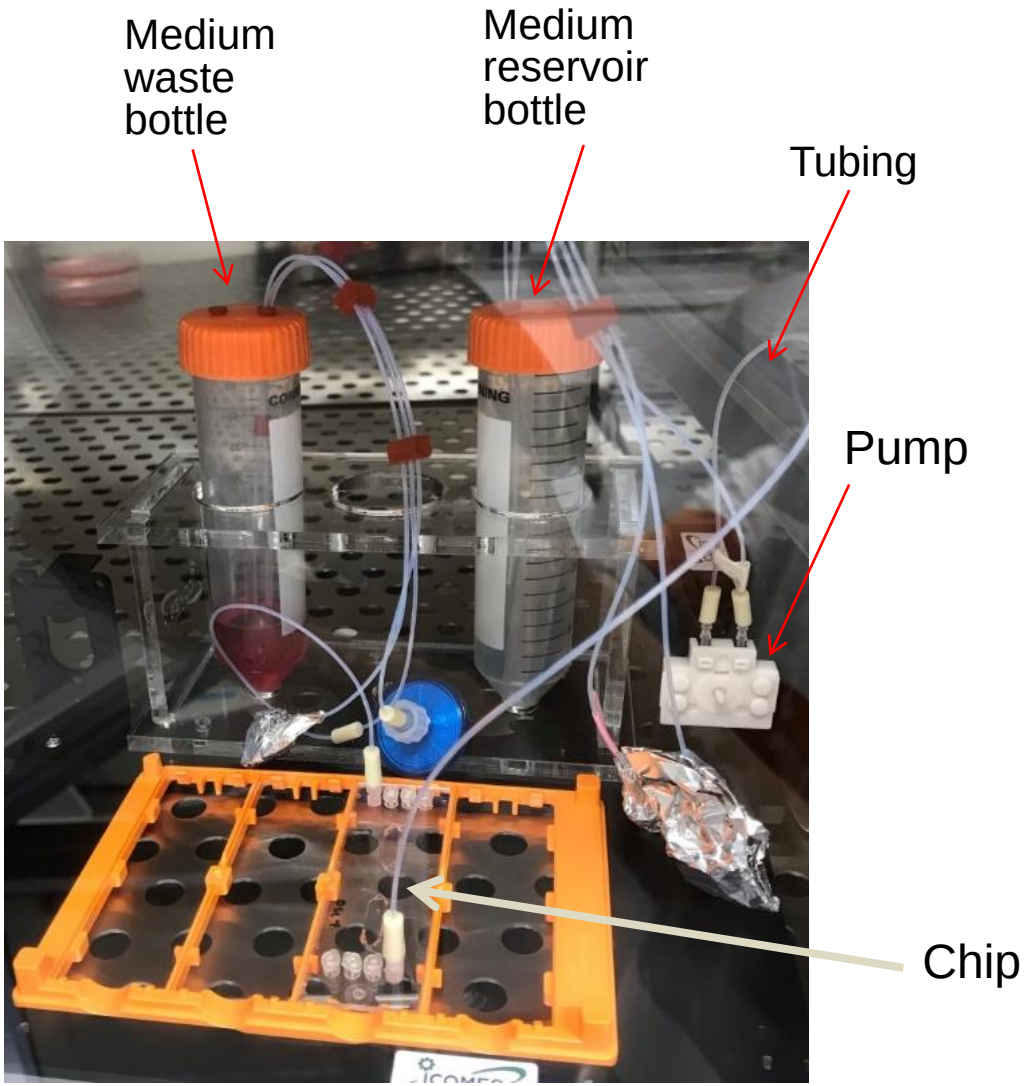
Challenge of Standardization in the AMED-MPS Project

1. Development Stage of Microphysiological System (MPS)

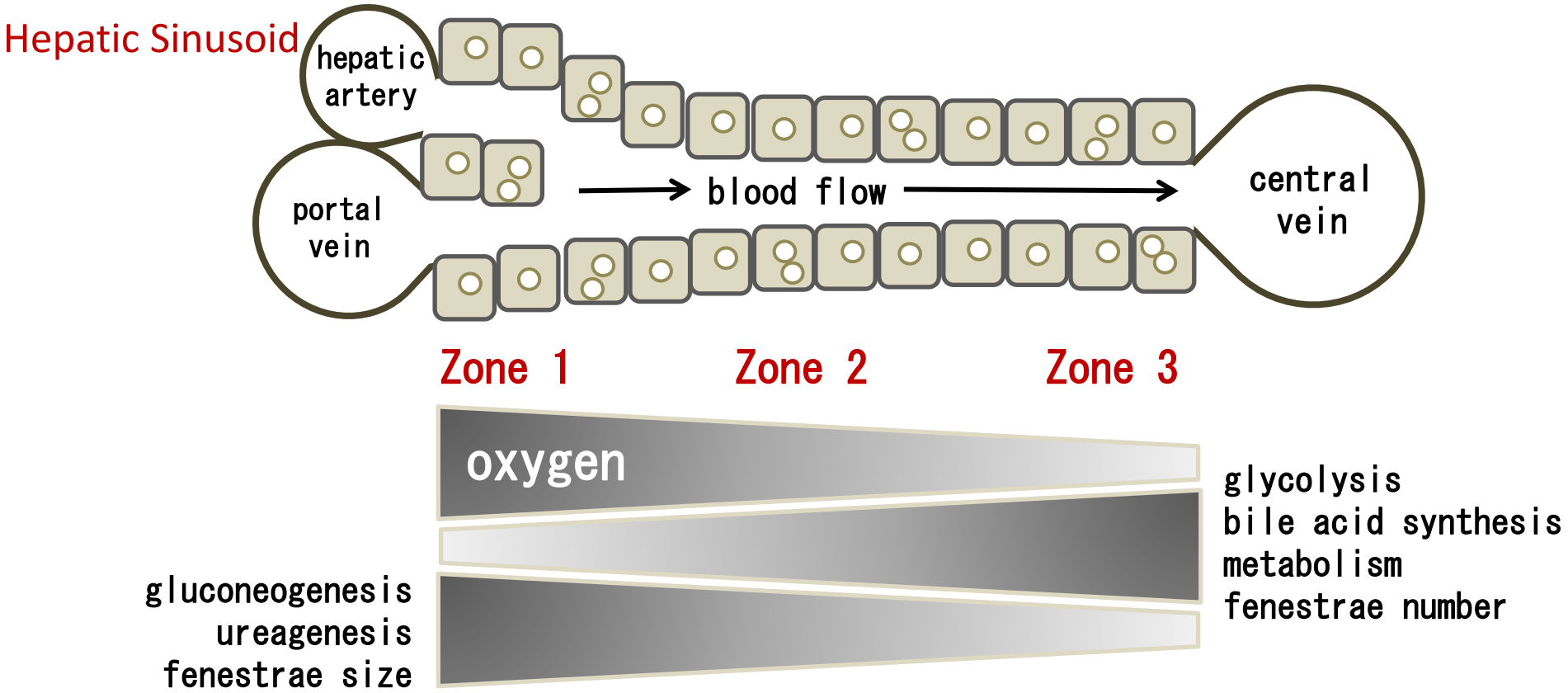
2. Points to Consider for Industrial Implementation of MPS

Development Stage of Microphysiological System

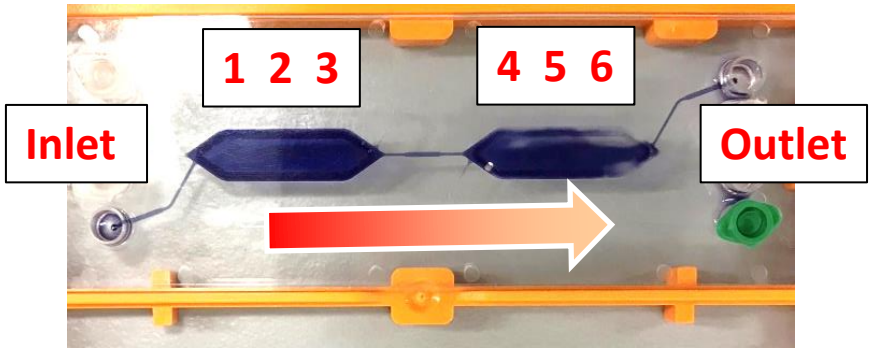
Assemble-type MPS

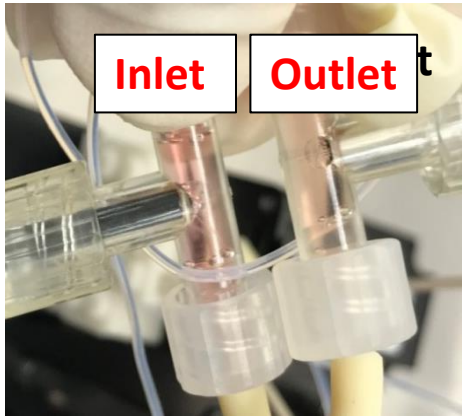


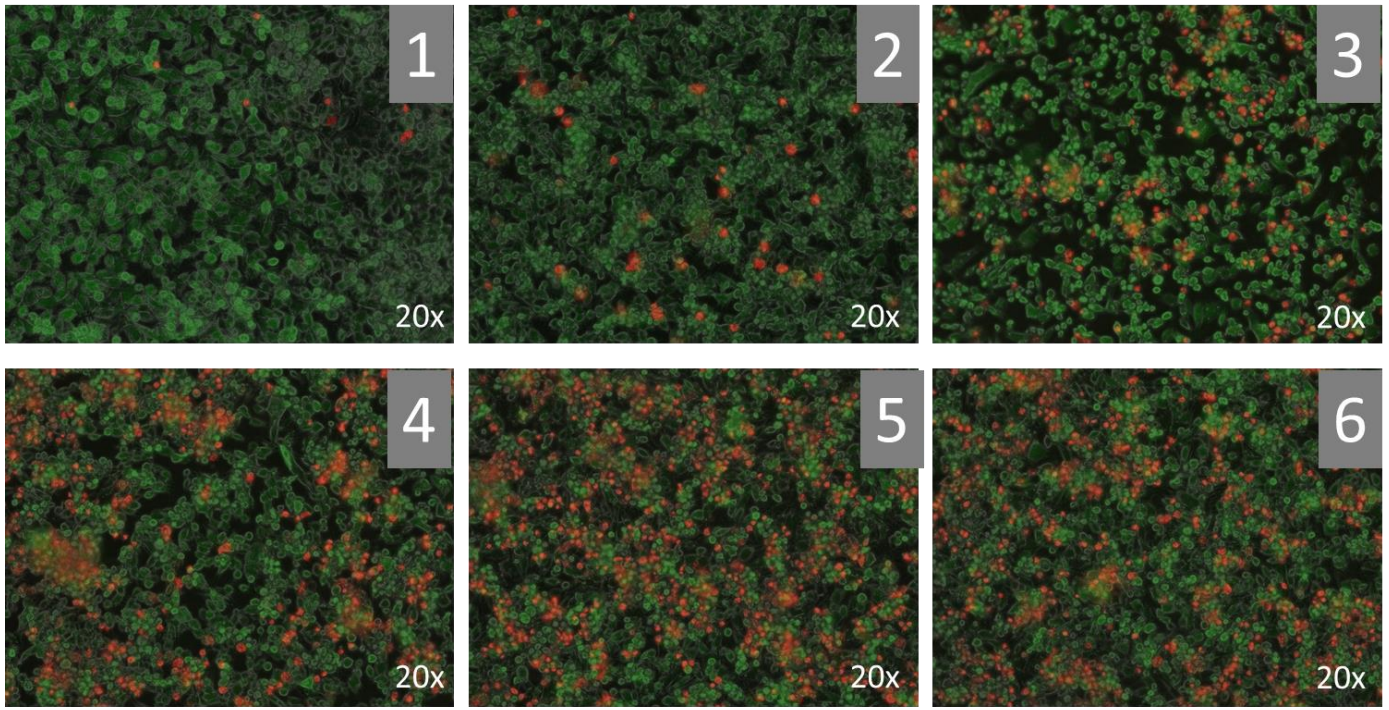
Hepatic Zonation and Region-specific Functional Expression



Gradient Formation according to Medium Flow

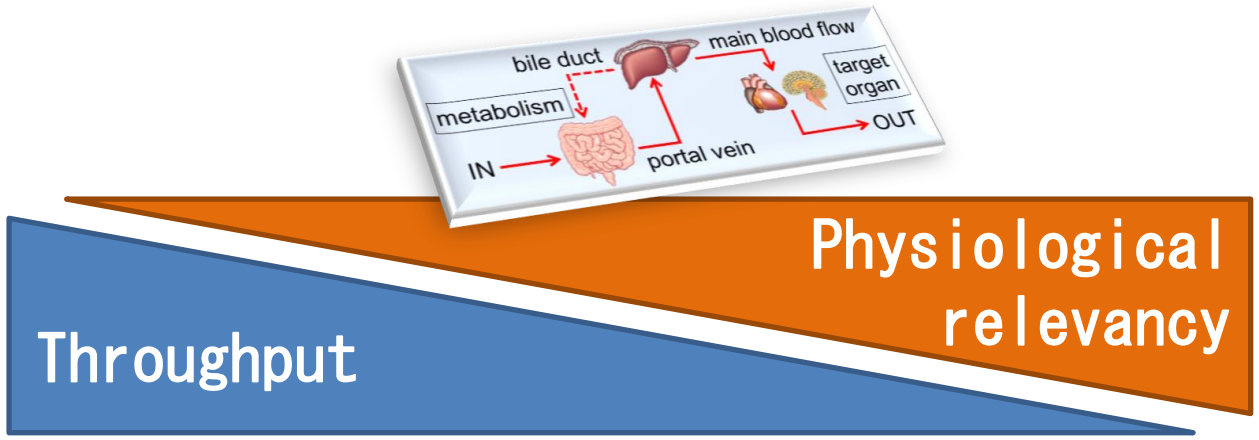






LIVE/DEAD staining（20x）

Availability and Application of MPS for *in vitro* Test



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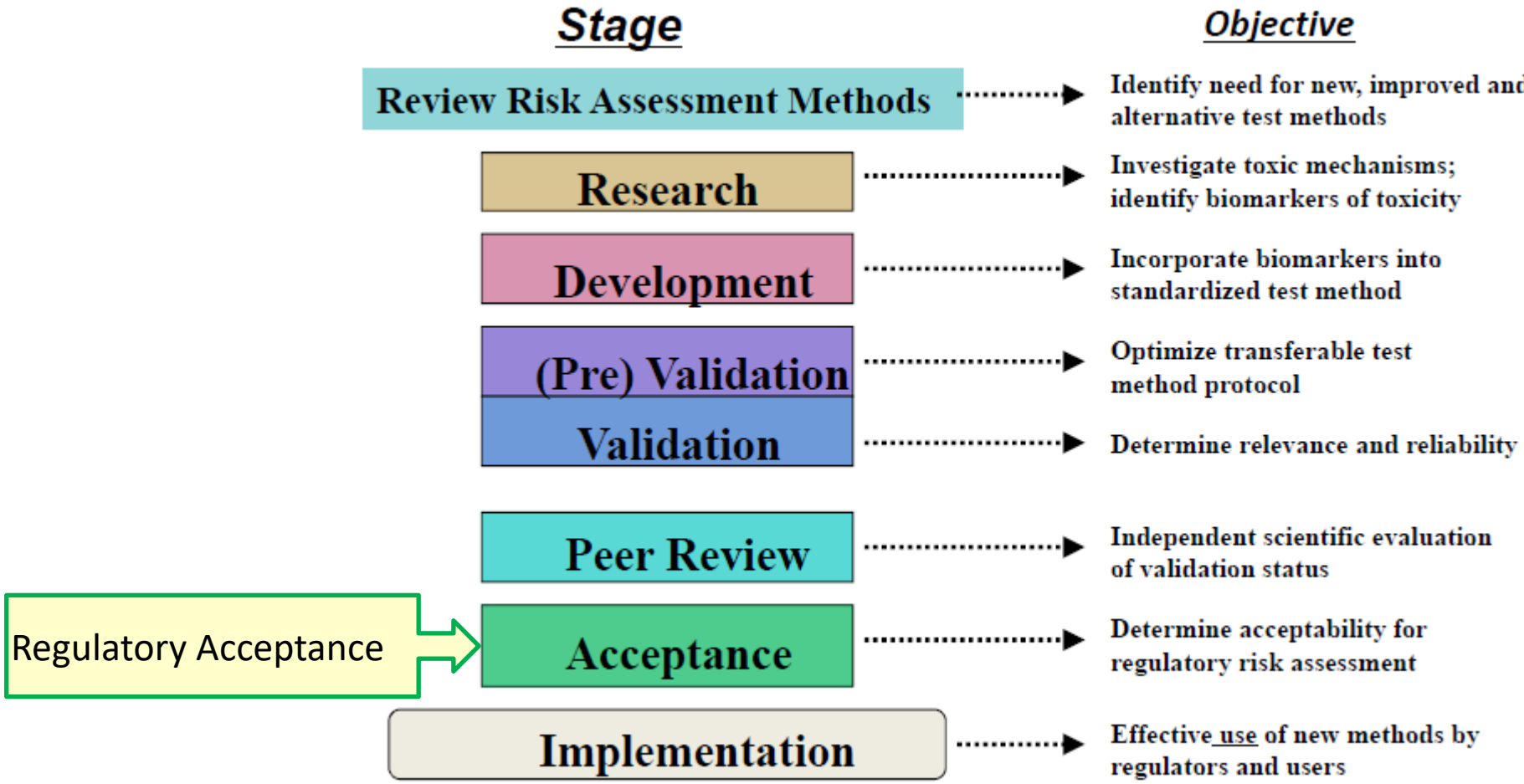
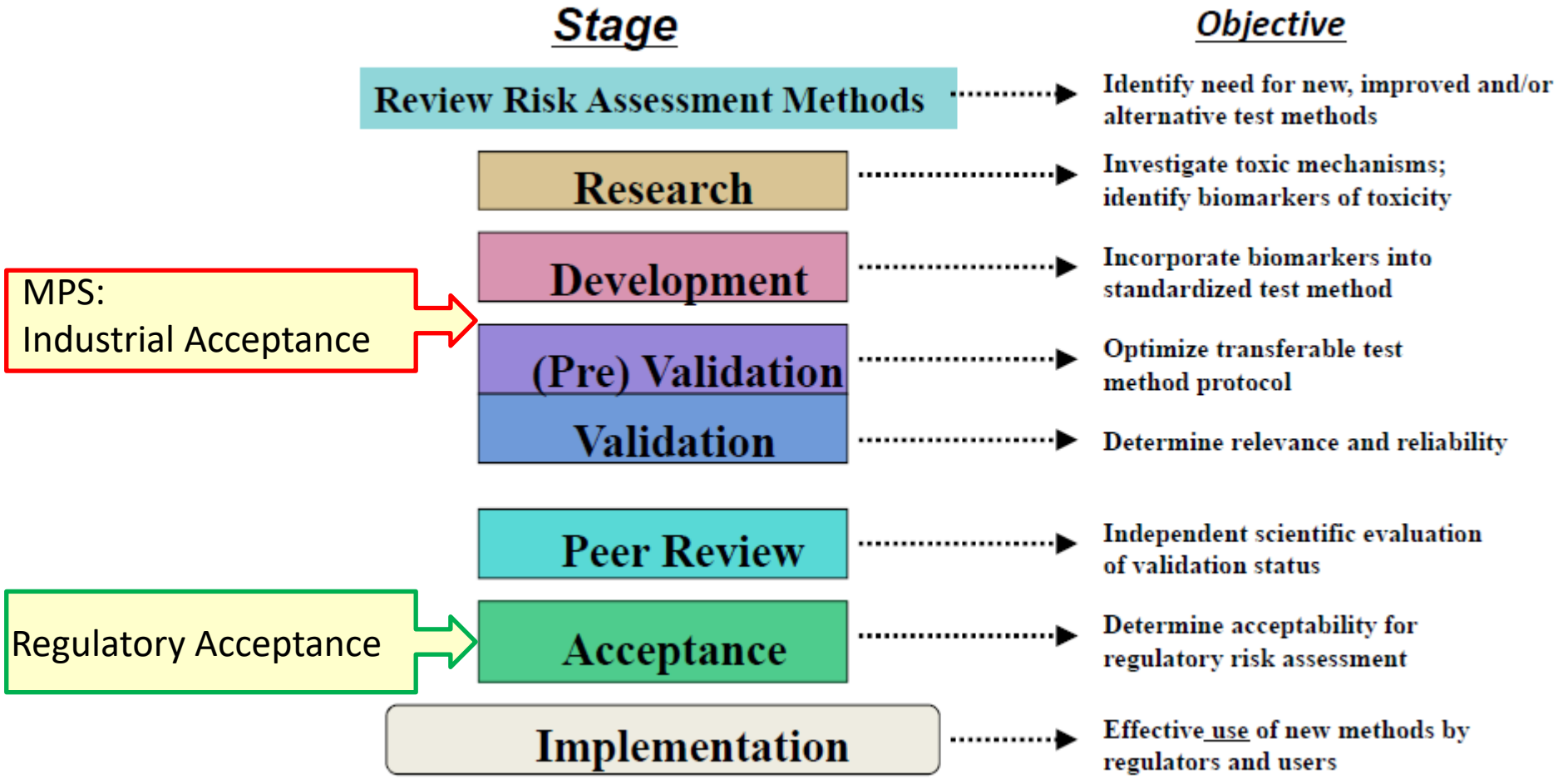
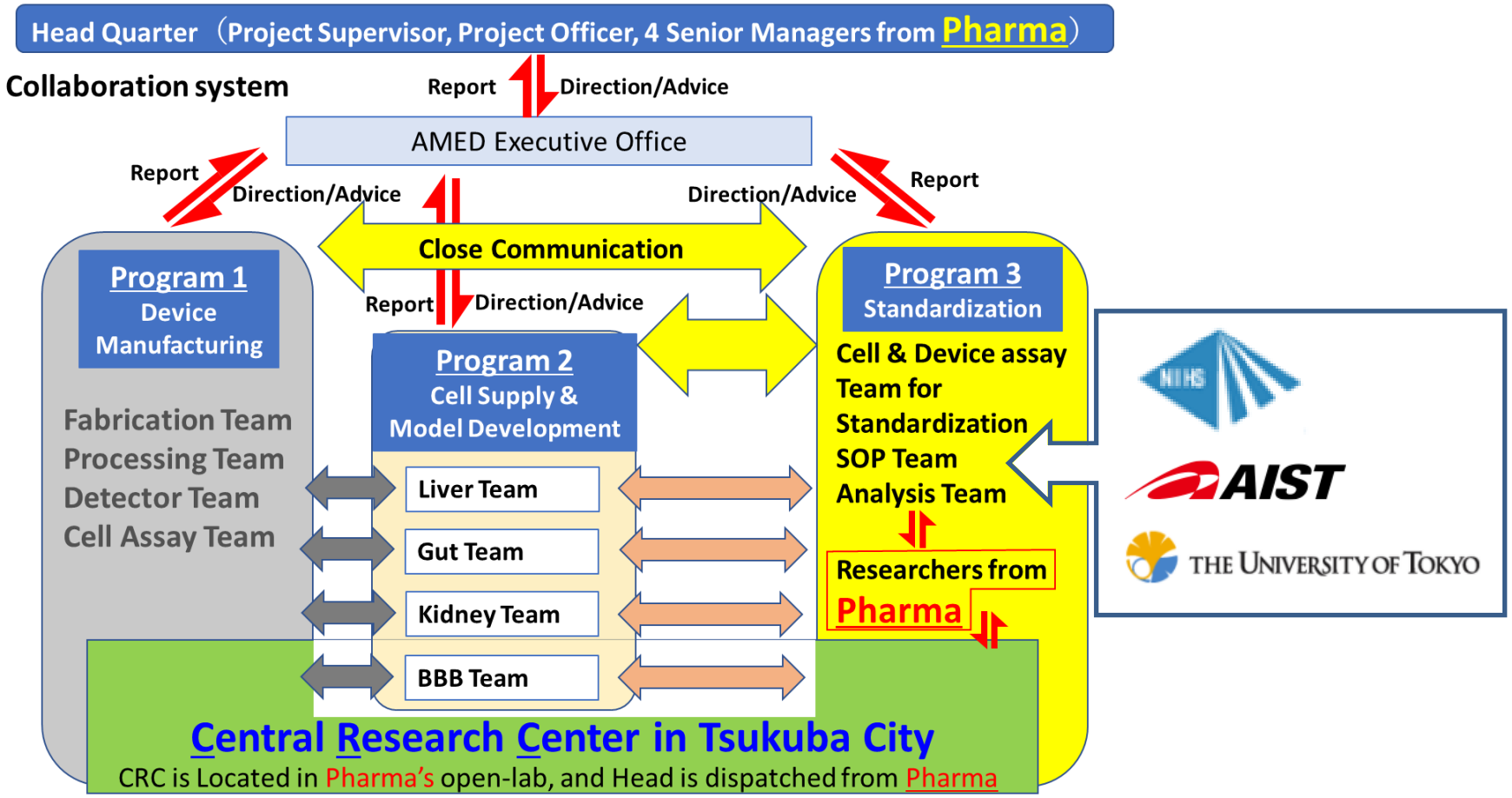
Test Method Evolution and Translation Process: Concept to Implementation



Test Method Evolution and Translation Process: Development Stage of MPS



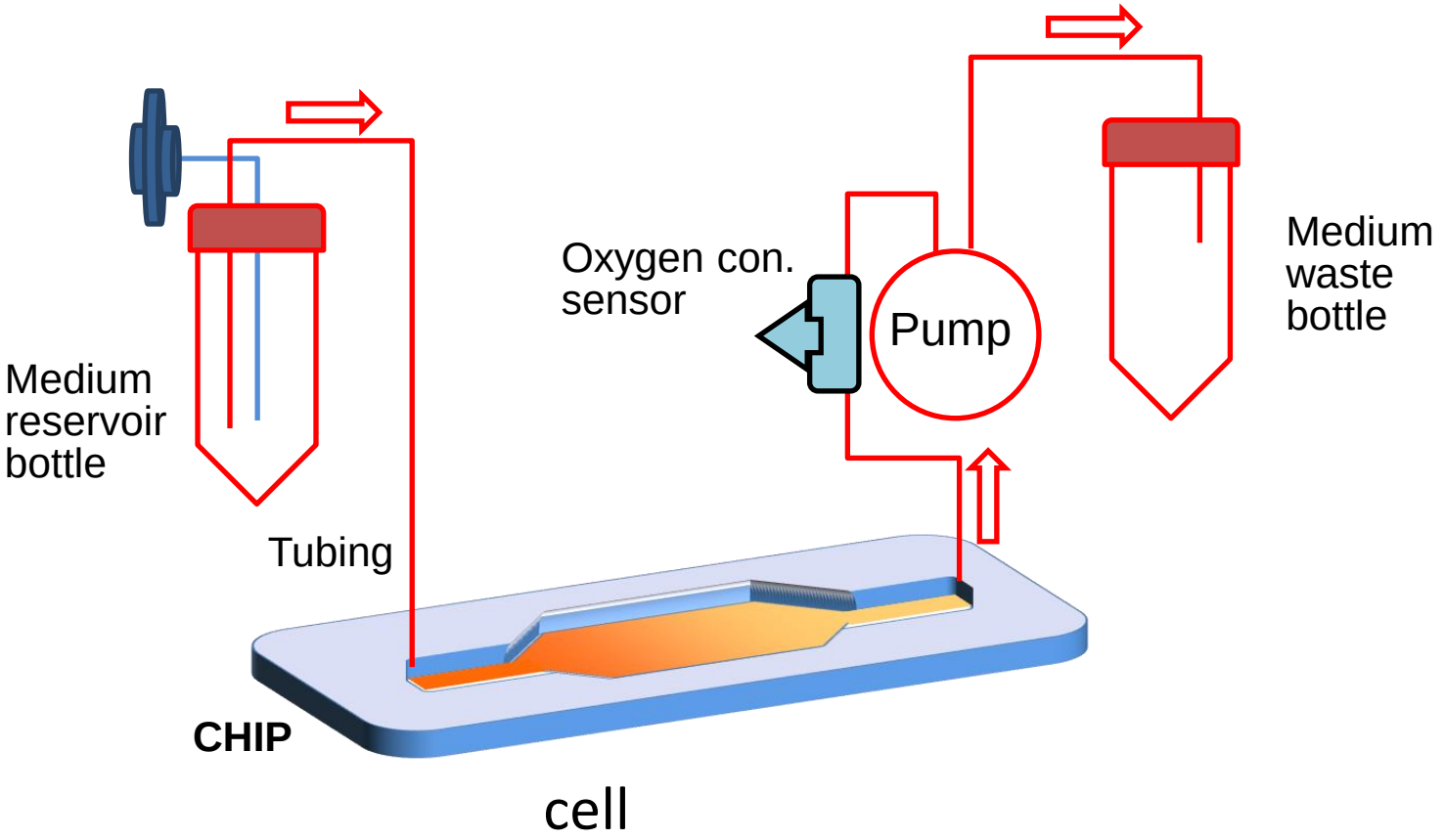
Challenge of Standardization in the AMED-MPS Project



- AMED (Japan Agency for Medical Research and Development) governed by the Cabinet Office.
- Targeting organs are liver, intestine, kidney and BBB.
- 30 organizations with ca. 130 researchers and engineers under one intellectual property agreement.
- Ca. 700 mil Yen/year from 2017 FY to 2021 FY.

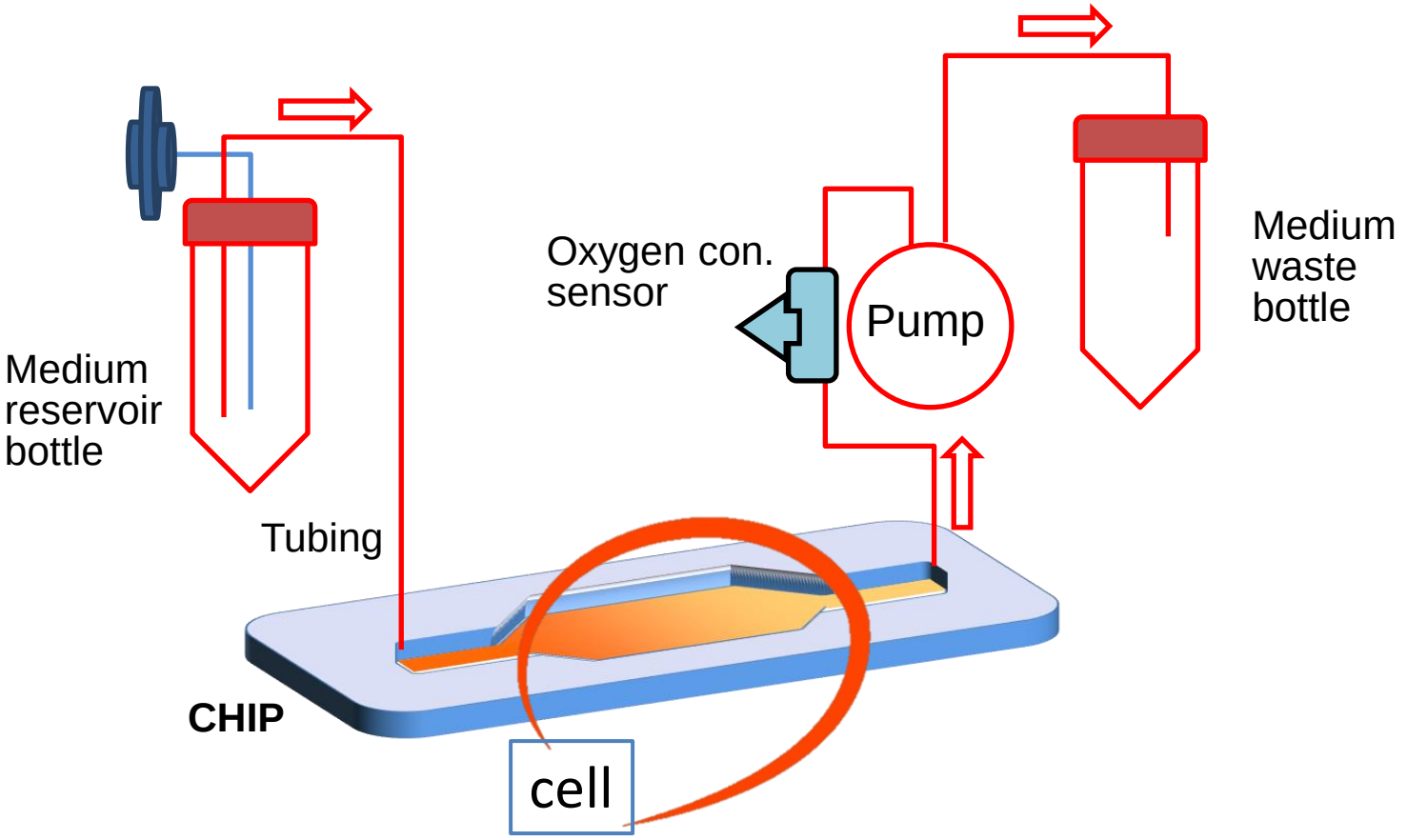
Points to Consider for Industrial Implementation of MPS

Points for MPS Performance Criteria



Points for MPS Performance Criteria

- Cell -



Minimal Requirements for liver-MPS

Tissue	Standard existing evaluation system	Required profile	Evaluation target	Measurement item
Liver	human cryo-preseved hepatocyte	・ has sufficient drug metabolic activity.	Expression of phase I enzyme activity	CYP、AO、FMO、MAO、CES
			Expression of phase II enzyme activity	UGT、SULT、GST
		・ has sufficient transporter activity.	Functional expression of transporter	ABC、SLC
		・ has the ability to induce the drug metabolizing enzymes.	induction of CYPs	CYP1A2, CYP2B6, CYP3A4、nuclear receptor
		・ capable of long-term culture.	cellular function	MTT、albumin、urea metabolism
		・ The structure of a micro bile duct can be confirmed.	Bile pocket formation	Localization of the biliary transporter Bile excretion capability
		・ has the ability to excrete bile.	Biliary transporter expression	BSEP, MRP2, BCRP, PGP
		・ Long-term repeated exposure that mimics the living body	Zonation	Functional gradient
		・ Covering various toxicity mechanisms	Liver fibrosis	aSMA, collagen

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		· Long-term repeated exposure that mimics the living body	Zonation	Functional gradient
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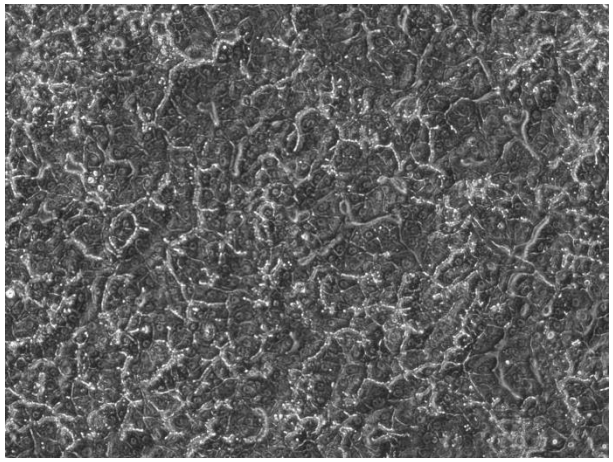
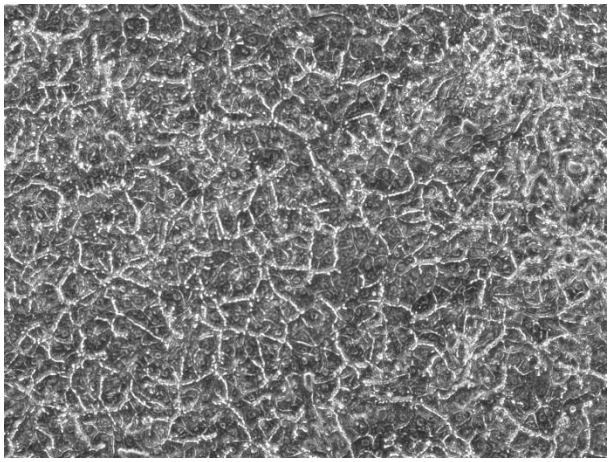
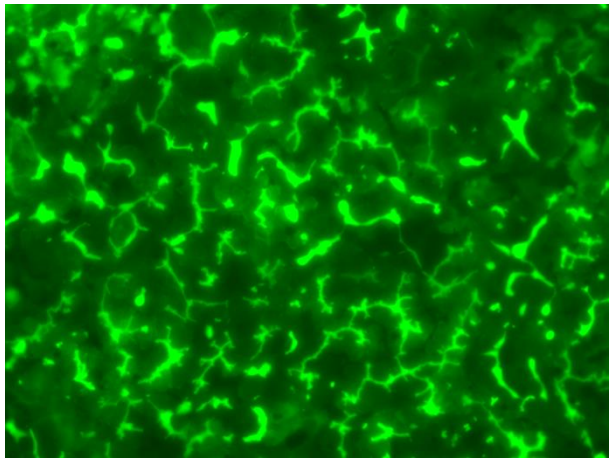
Bile Canaliculi Formation

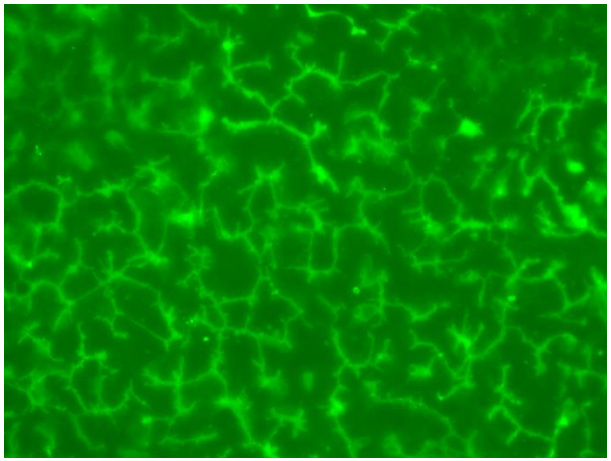
iPSC-hep: long term culture + sandwich culture ×20



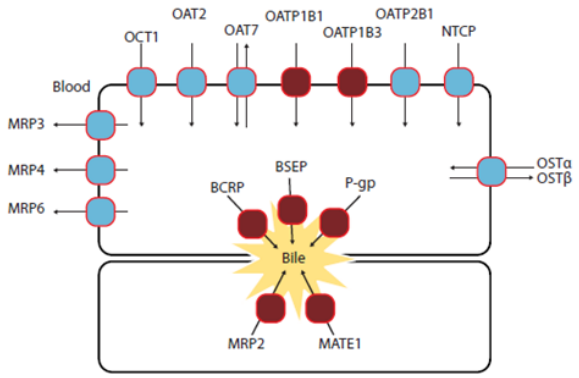




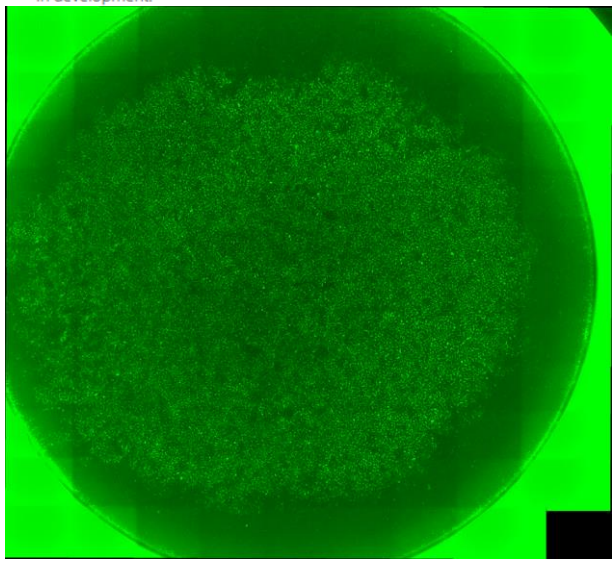
MRP2
(Fluorescein diacetate)



BSEP
(Tauro-nor-THCA-24DBD)



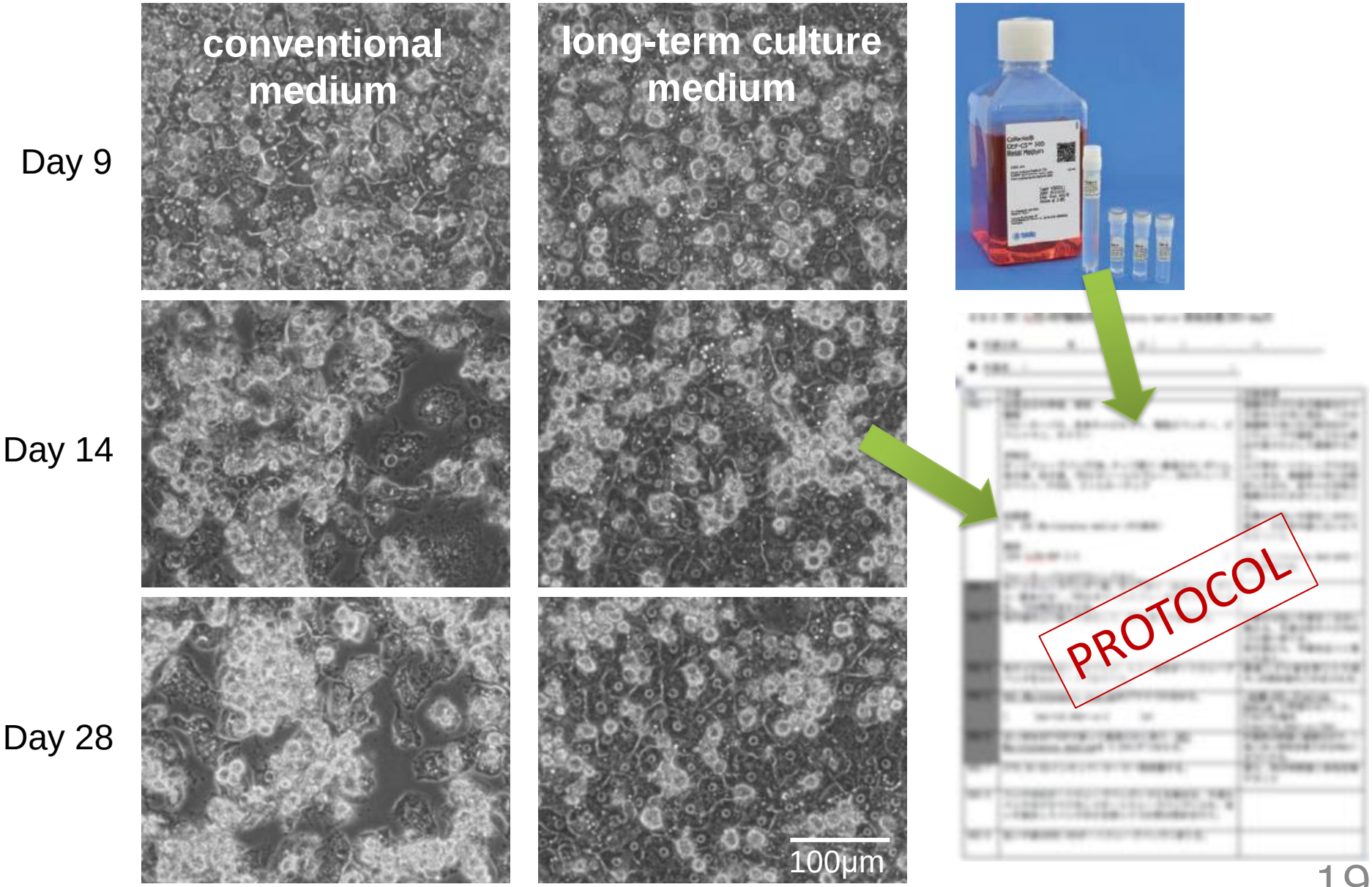
Common uptake and efflux transporters in human liver
Highlighted (red) transporters represent knockout cell lines currently in development.



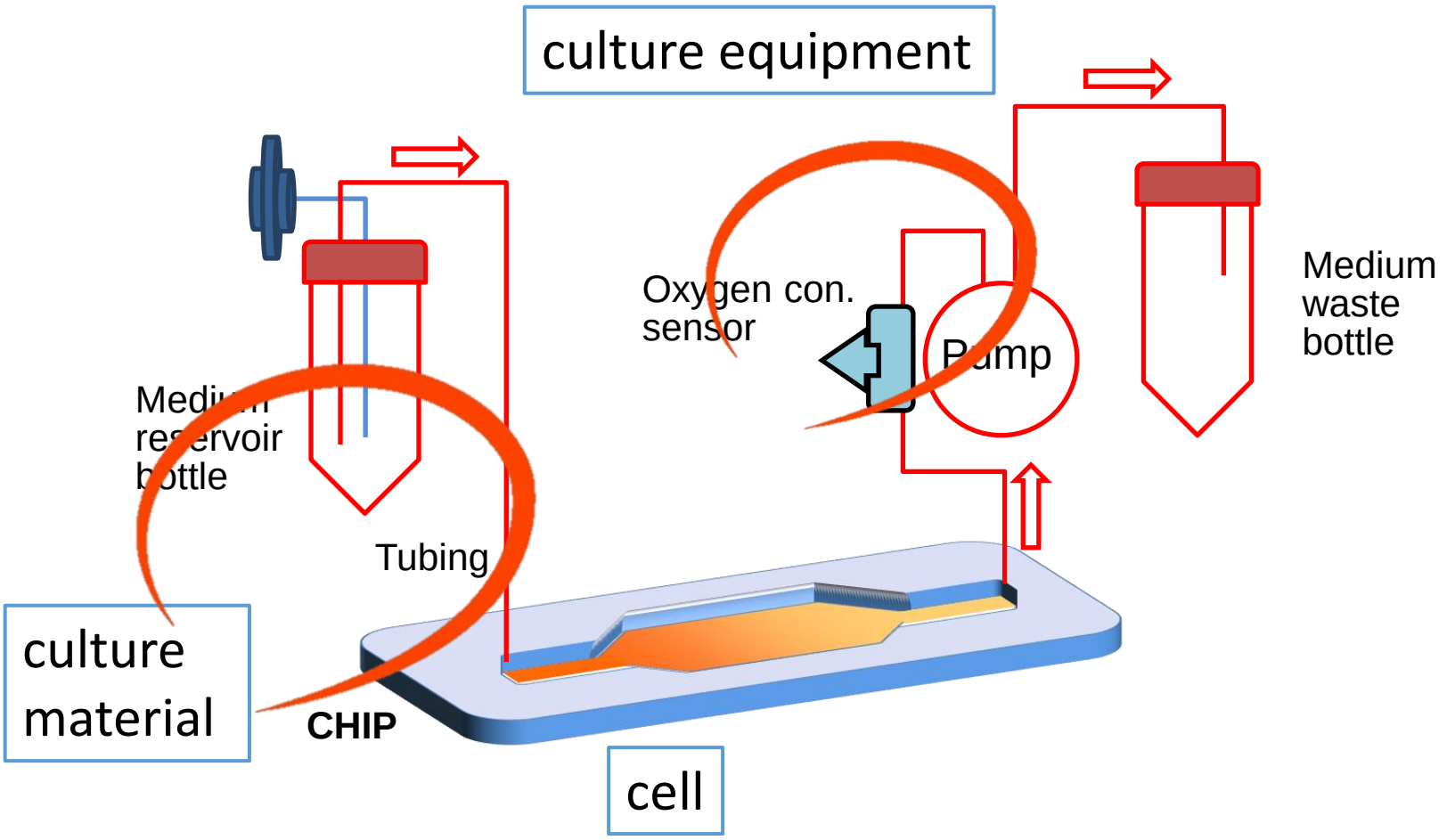
Overall observation of
the culture-well bottom

Fluorescent substrates of MRP2 and BSEP were excreted into the bile canaiiculi.

Cell, Medium, and Protocol



Points for MPS Performance Criteria
- Material & Equipment -



Points for MPS Performance Criteria
- Material & Equipment -

Examples of consideration points…

1. Sterilization (radiation, autoclave, and gas sterilization) was necessary, but the sterilization process caused deterioration of the characteristics of the equipment.
2. Chemical substances such as solvents used in the laminating of the substrate affected the cell culture.
3. Autofluorescence of the cell culture substrate prevented cell observation.
4. Adsorption of the fluorescent reagent used for cell observation occurred and observation was not possible.
5. … … …

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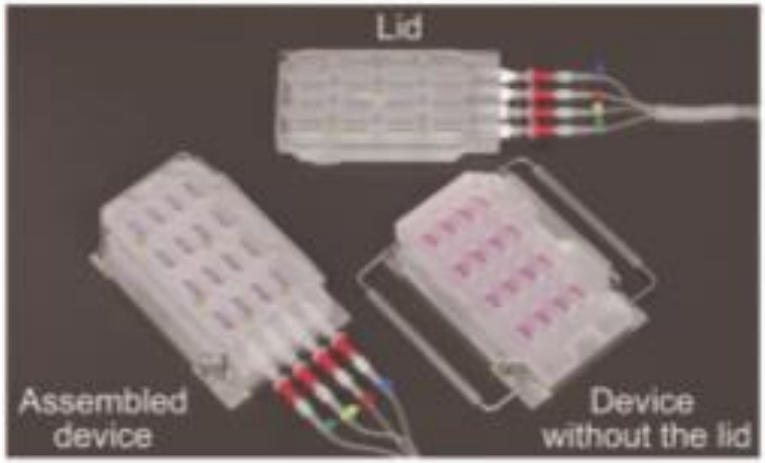
A Solution for Sterilization: Pressure Driven-MPS

A multi-throughput multi-organ-on-a-plate on a pneumatic pressure-driven medium circulation platform †

T. Satoh,[a‡] S. Sugiura,[a‡] K. Shin,[a] R. Onuki-Nagasaki,[a] S. Ishida,[b] K. Kikuchi,[c] M. Kakiki,[c] and T. Kanamori [a]



- microplate-sized pneumatic pressure-driven multi-organ culture platform
- pneumatic pressure directly drives the liquid.
- connections to the pneumati pressure lines are easily detachable.
- lid is easily removed.

Points for MPS Performance Criteria
- Material & Equipment -

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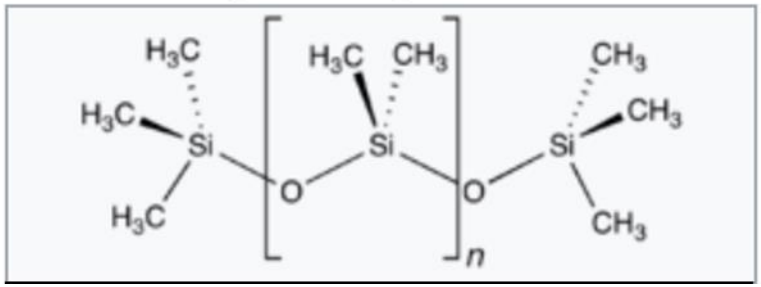
PDMS

Polydimethylsiloxane



cmgh
CELLULAR AND MOLECULAR GASTROENTEROLOGY AND HEPATOLOGY

REVIEW

Microfluidic Organ-on-a-Chip Models of Human Intestine

Amir Bein,[1,a] Woojung Shin,[2,a] Sasan Jalili-Firoozinezhad,[1,3] Min Hee Park,[2] Alexandra Sontheimer-Phelps,[1,4] Alessio Tovaglieri,[1,5] Angeliki Chalkiadaki,[1] Hyun Jung Kim,[2] and Donald E. Ingber[1,6,7]

The Gut Chip is made of a flexible, gas-permeable, silicone polymer (polydimethylsiloxane [PDMS]) that is crystal clear so that it allows high-resolution imaging by phase contrast, differential interference contrast, or immunofluorescence confocal microscopy.

……

Although PDMS holds many favorable properties for manufacturing microfluidic organ devices, it also has the potential drawback of adsorbing small and hydrophobic molecules.

Organs-on-chips are often fabricated in part or wholly from polydimethylsiloxane (PDMS), an oxygen-permeable, optically-clear, non-flammable, non-toxic silicon-based organic polymer. However, PDMS absorbs or binds compounds or proteins under certain conditions, leading to loss of drugs or compounds that are introduced into the system. This is undesirable in the context of organs-on-chips as it reduces the ability to accurately assess protein binding or calculate dosage ranges and responses of small molecules. [https://www.sbir.gov/sbirsearch/detail/1508473]

A Solution for Adsorption: PDMS-free MPS

micromachines

京都大学 KYOTO UNIVERSITY AGC

Article

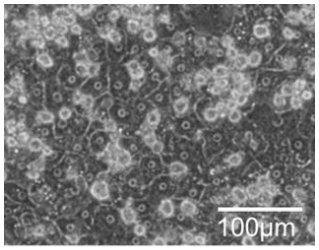
Tetrafluoroethylene-Propylene Elastomer for Fabrication of Microfluidic Organs-on-Chips Resistant to Drug Absorption

Emi Sano [1,†], Chihiro Mori [1,†], Naoki Matsuoka [2], Yuka Ozaki [1], Keisuke Yagi [2], Aya Wada [2], Koichi Tashima [2], Shinsuke Yamasaki [2], Kana Tanabe [2], Kayo Yano [1] and Yu-suke Torisawa [1,3,*]

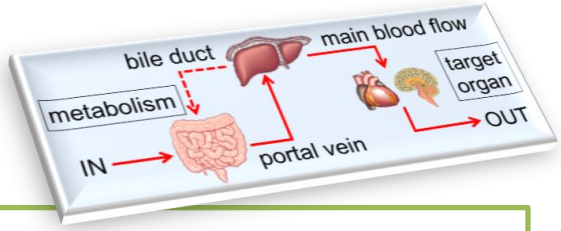
Absorption of rhodamine B into PDMS and FEPM.

Time-course analysis of drug absorption into PDMS, FEPM, and polystyrene.

Challenge of Standardization in the AMED-MPS Project

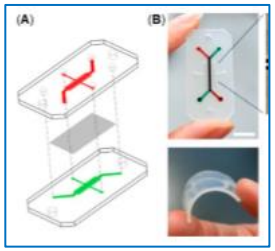


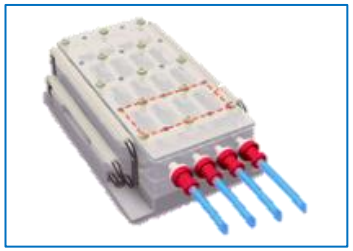
Cell Criteria



MPS performance standard

Standard for material and equipment





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- Research Project for Practical Applications of Regenerative Medicine
- Research on Development of New Drugs.