# IQ MPS Affiliate Perspective on Characteristics and Requirements for New Approach Methodologies (NAMs)

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| | | | | |
|---|---|---|---|---|
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| Amgen | BMS | Janssen | Sanofi | Theravance |
| Alnylam | Eisai | Merck | Seattle Genetics | Vertex |
| Astellas | Eli Lilly | Merck Healthcare KGaA | | |
| AstraZeneca | Genentech | Novartis | | |

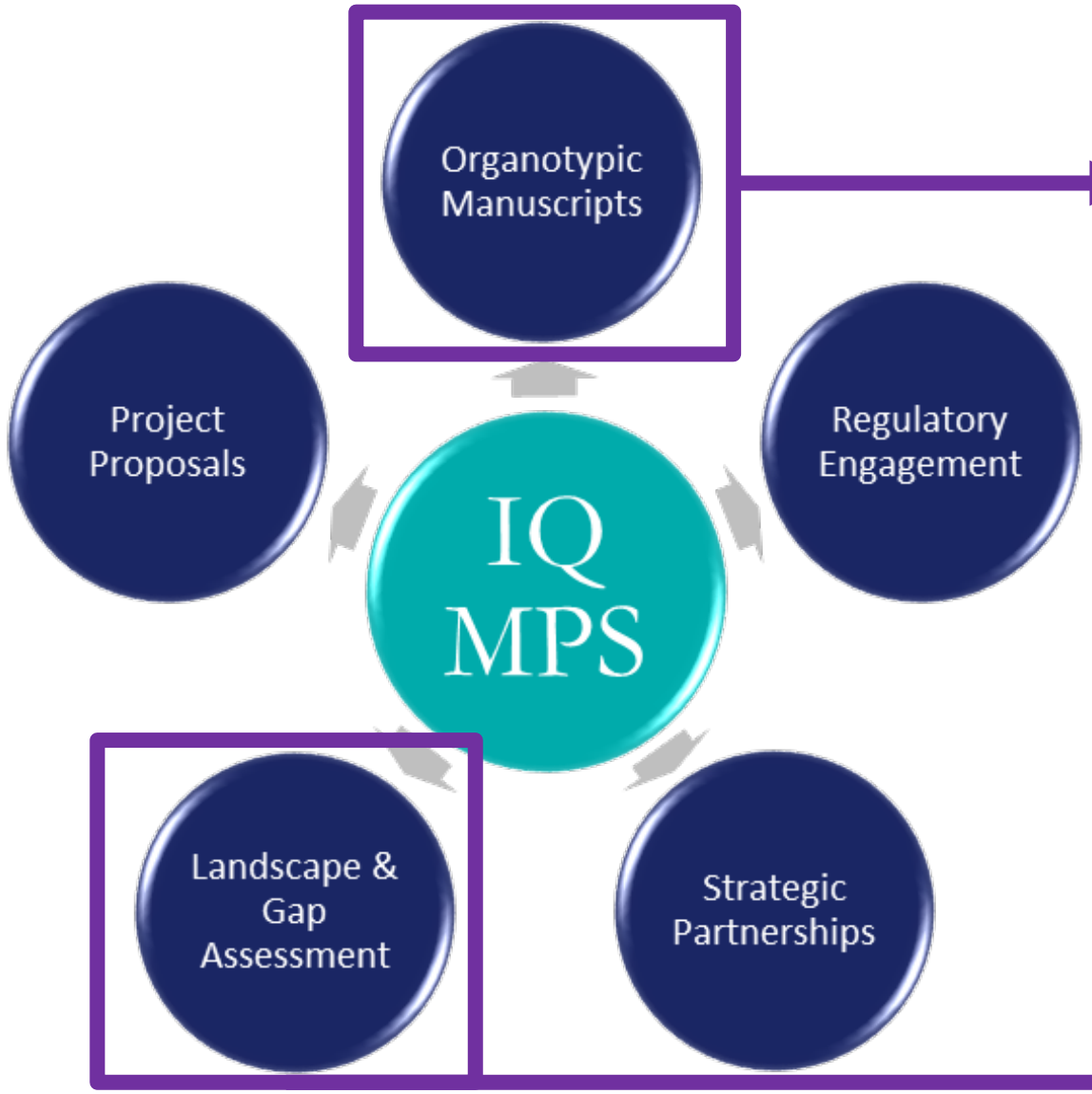
Serve as a thought leader for both MPS developers and stakeholder organizations in the industry implementation and qualification of MPS models.

Provide a venue for appropriate cross-pharma collaboration and data sharing to facilitate industry implementation and qualification of MPS models.

Create focused engagement between industry and regulatory agencies on the current status and evolving field of MPS in an industry setting.

Develop external partnerships and collaborations to help enhance the inclusion of industry priorities.

# Current Working Groups




- Challenges/gaps in current *in vitro* and *in vivo* models
- Data and capabilities needed to demonstrate a meaningful improvement over standard 2D systems
- Industry suggested endpoints, online readouts, test compounds, etc. for “MPS qualification” studies

- Understand how pharmaceutical companies are currently using complex *in vitro* models (CIVM) and what are the major challenges to realizing the full potential of this technology in the context of drug research and development

# Organotypic Manuscripts

Lab on a Chip

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Cite this: *Lab Chip*, 2020, **20**, 1049

## Introduction to a manuscript series on the characterization and use of microphysiological systems (MPS) in pharmaceutical safety and ADME applications

Kristin Fabre,[ab] Brian Berridge,[c] William R. Proctor,[d] Sherry Ralston,[e] Yvonne Will,[f] Szczepan W. Baran,[g] Gorm Yoder[h] and Terry R. Van Vleet *[e]

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Cite this: *Lab Chip*, 2020, **20**, 215

## Liver microphysiological systems development guidelines for safety risk assessment in the pharmaceutical industry

Andreas R. Baudy, *[a] Monicah A. Otieno,[b] Philip Hewitt,[c] Jinping Gan,[d] Adrian Roth,[e] Douglas Keller,[f] Radhakrishna Sura,[g] Terry R. Van Vleet[g] and William R. Proctor[h]

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Cite this: *Lab Chip*, 2020, **20**, 199

## Drug-induced skin toxicity: gaps in preclinical testing cascade as opportunities for complex *in vitro* models and assays

Rhiannon N. Hardwick, *[a] Catherine J. Betts,[b] Jessica Whritenour,[c] Radhakrishna Sura,[d] Maike Thamsen,[e] Elad H. Kaufman[f] and Kristin Fabre†[g]

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Cite this: *Lab Chip*, 2019, **19**, 3152

## Microphysiological lung models to evaluate the safety of new pharmaceutical modalities: a biopharmaceutical perspective

Garrett R. Ainslie, *[a] Myrtle Davis,[b] Lorna Ewart,[c] Linda A. Lieberman,[d] David J. Rowlands,[e] Andrew J. Thorley, [e] Gorm Yoder[f] and Anne M. Ryan[g]

dates

20, **20**, 468

## A pharmaceutical industry perspective on microphysiological kidney systems for evaluation of safety for new therapies

Jonathan A. Phillips, [a] Taraka Sai Pavan Grandhi,[b] Myrtle Davis,[c] Jean-Charles Gautier,[d] Niresh Hariparsad,[a] Douglas Keller, [e] Radhakrishna Sura[f] and Terry R. Van Vleet*[f]

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Cite this: *Lab Chip*, 2020, **20**, 1177

## Developing *in vitro* assays to transform gastrointestinal safety assessment: potential for microphysiological systems†

Matthew F. Peters, *[a] Allison L. Choy,[b] Carmen Pin,[c] Derek J. Leishman,[d] Annie Moisan,[e] Lorna Ewart, ‡[a] Peggy J. Guzzie-Peck,[f] Radhakrishna Sura,[g] Douglas A. Keller, [h] Clay W Scott[a] and Kyle L. Kolaja [i]

ates

, **20**, 446

## Microphysiological systems for ADME-related applications: current status and recommendations for system development and characterization

Stephen Fowler, [a] Wen Li Kelly Chen,[b] David B. Duignan,[c] Anshul Gupta,[b] Niresh Hariparsad,[d] Jane R. Kenny,[e] W. George Lai,[f] Jennifer Liras, [g] Jonathan A. Phillips [h] and Jinping Gan *[i]

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Cite this: *Lab Chip*, 2020, **20**, 697

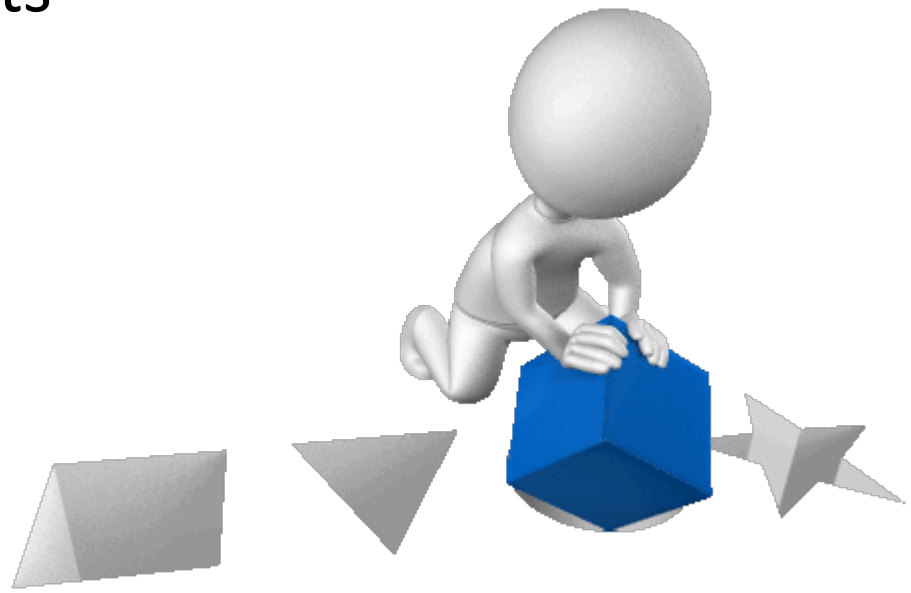
## Application of microphysiological systems in biopharmaceutical research and development

Norman C. Peterson, *[a] Prathap Kumar Mahalingaiah,[b] Aaron Fullerton[c] and Matteo Di Piazza[d]

https://www.iqmps.org/publications

# Defining the Problem

- Models for predicting adverse human events
- Prediction of human toxicities
  - Rodents 43%
  - Non-rodents 63%
- Highest concordance
  - Hematological, gastrointestinal and cardiovascular toxicity
- Lowest concordance
  - Musculoskeletal
  - Respiratory
  - Neurological
  - Hepatic

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- C. Tamaki, T. Nagayama, M. Hashiba, M. Fujiyoshi, M. Hizue, H. Kodaira, M. Nishida, K. Suzuki, Y. Takashima, Y. Ogino, D. Yasugi, Y. Yoneta, S. Hisada, T. Ohkura and K. Nakamura, *J. Toxicol. Sci.*, 2013, **38**, 581–598.
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# Definition of Complex *In Vitro* Models (CIVM)

"Going beyond traditional 2D culture by including several of the following design aspects:

- multi-cellular environment within biopolymer or tissue-derived matrix;
- 3D structure;
- inclusion of mechanical cues such as stretch or perfusion for breathing, gut peristalsis, flow;
- incorporate primary or stem cell derived cells;
- and/or inclusion of immune system components."

L. Ewart, K. Fabre, A. Chakilam, Y. Dragan, D. B. Duignan, J. Eswaraka, J. Gan, P. Guzzie-Peck, M. Otieno, C. G. Jeong, D. A. Keller, S. M. de Morais, J. A. Phillips, W. Proctor, R. Sura, T. Van Vleet, D. Watson, Y. Will, D. Tagle and B. Berridge, **Navigating tissue chips from development to dissemination: A pharmaceutical industry perspective.** *Exp. Biol. Med.*, 2017, **242**, 1579–1585

# Must be a Value Add

- Replace poor or non-existing models of toxicity

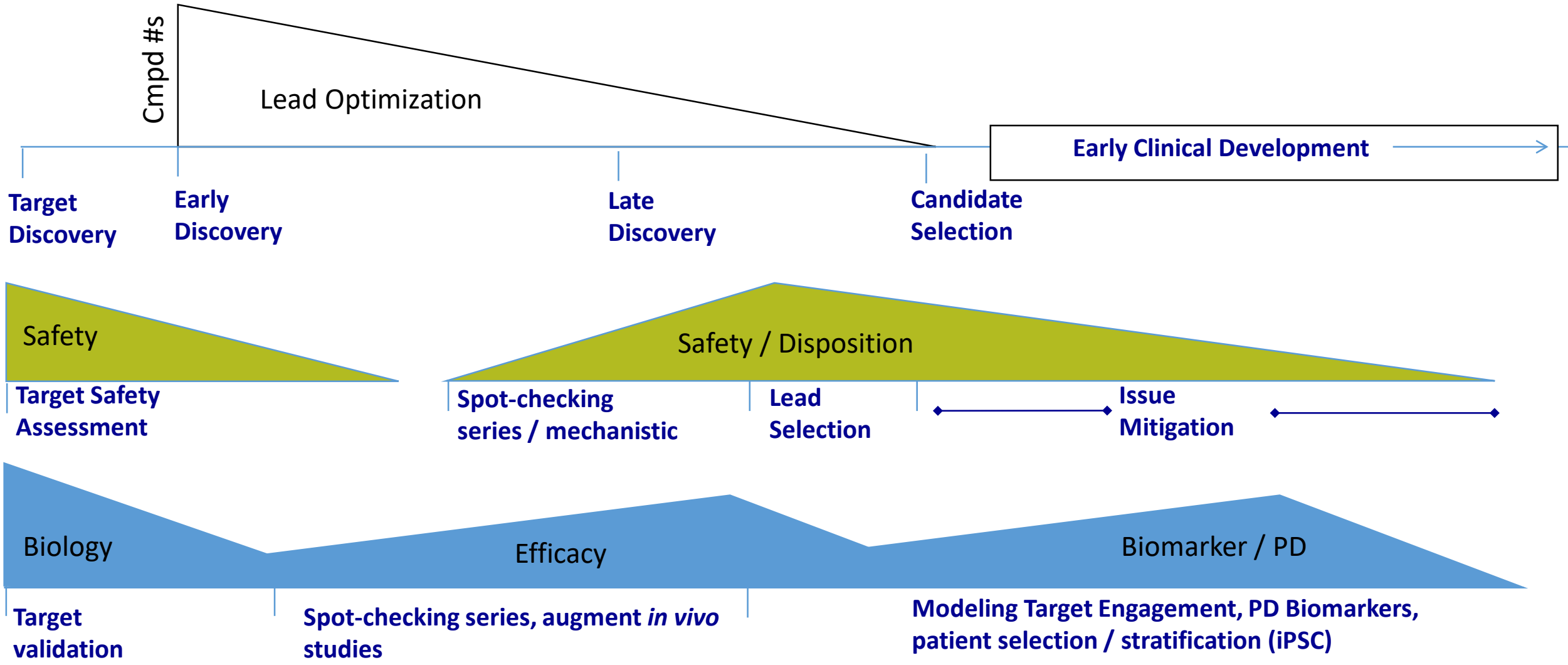
| Clinical/preclinical finding | Current models | Need |
|---|---|---|
| Cholestasis | DICI models | Model with organized bile ducts and measurable bile flow |
| Glomerulopathy | Podocyte, mesangial cell cultures | Model with organized slit diaphragm and measurable filtration function |
| Crystalluria | None | Models of urine concentration and effects of compounds prone to cause solids in the collecting ducts |
| Vascular injury | HUVEC, organ-specific primary endothelial cells | Organ specific architecture with organized vessels and appropriate interactions with tissue cells |
| Cardiac valvulopathy | None | Model of heart valves that responds properly to compounds causing valvulopathy |
| Neurodegeneration | Primary neurons, neurite outgrowth, iPSC-derived neurons | Models that replicate the complexity of nervous tissue with signalling transduction and properly respond to compounds causing effects after chronic exposures (*i.e.* >6 months). |
| Retinopathy | Retinal epithelium | Models that match the complexity of the retinal epithelium with various cell types and responds to prototypical functional alterations and retinopathy agents that disrupt vision |
| Inflammation/immune response | PBMC binding, cytokine release assays | Model(s) with appropriate immune components to reproduce the complexities of immune responses from drug treatment |

DICI = drug induced cholestatic index; HUVEC = human umbilical vein endothelial cells; PBMC = peripheral blood mononuclear cells.

- Show toxicities arising from previously undetectable mechanisms
- Identifying changes in cell function preceding adverse events

Fabre, K., Berridge, B., Proctor, W., Ralston, S., Will, Y., Baran, SW., Yoder, G., Van Vleet, T. (2019) **An Introduction to the Characterization and Use of Microphysiological Systems (MPS) in Pharmaceutical Safety and ADME Applications**. *Lab on a Chip*

# Context of Use: Right Model for Right Need

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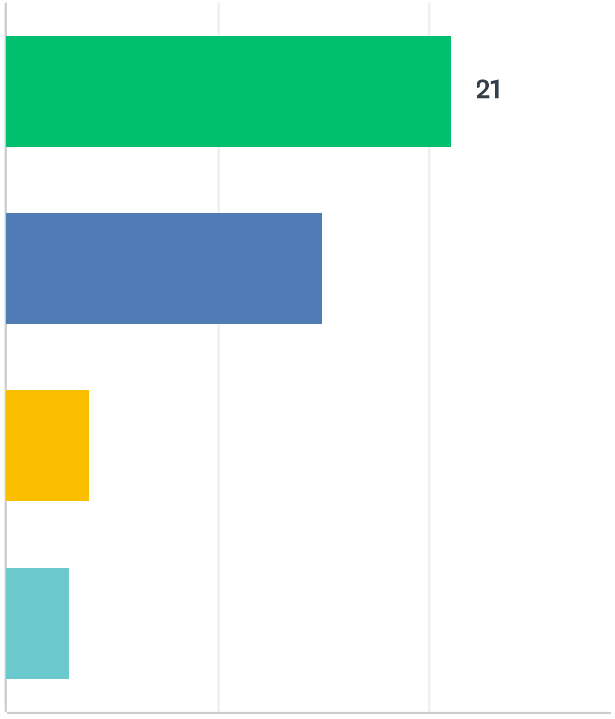
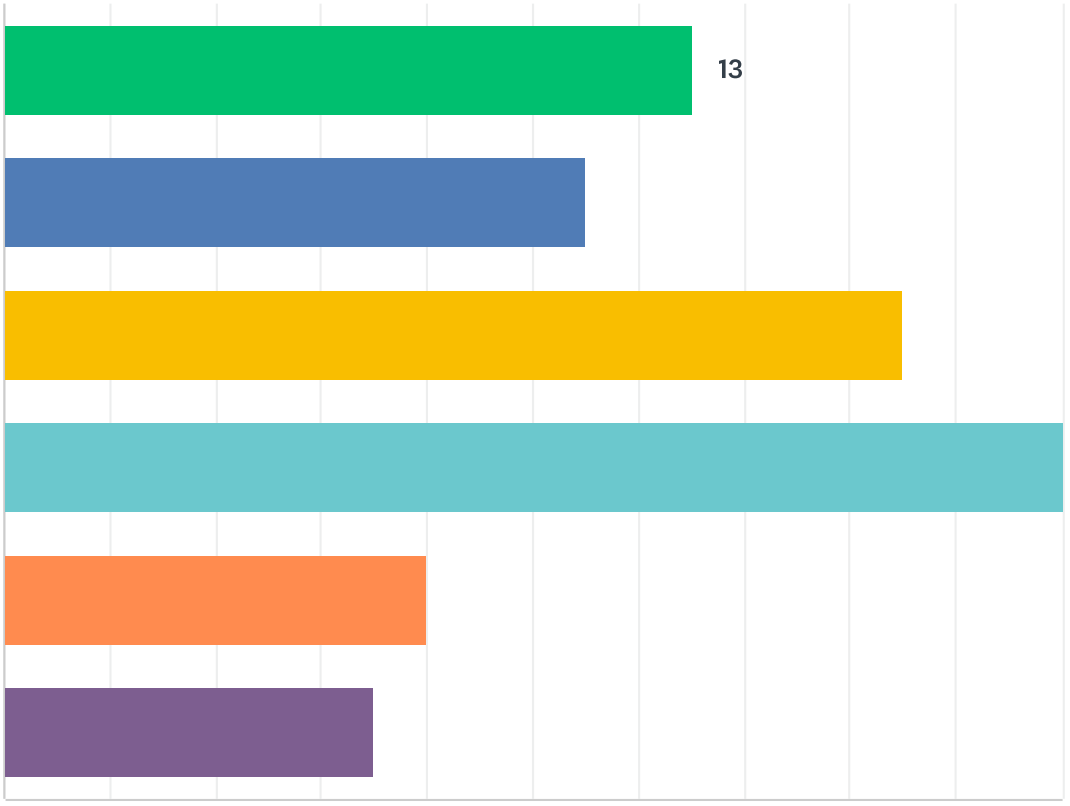
# Landscape Analysis / Gap Assessment Survey Preliminary Results

In general, what modalities are you testing using CIVM? Please select all that apply.




For which stage of drug discovery and development is your company currently evaluating and/or applying CIVM?

*Please select all that apply.*

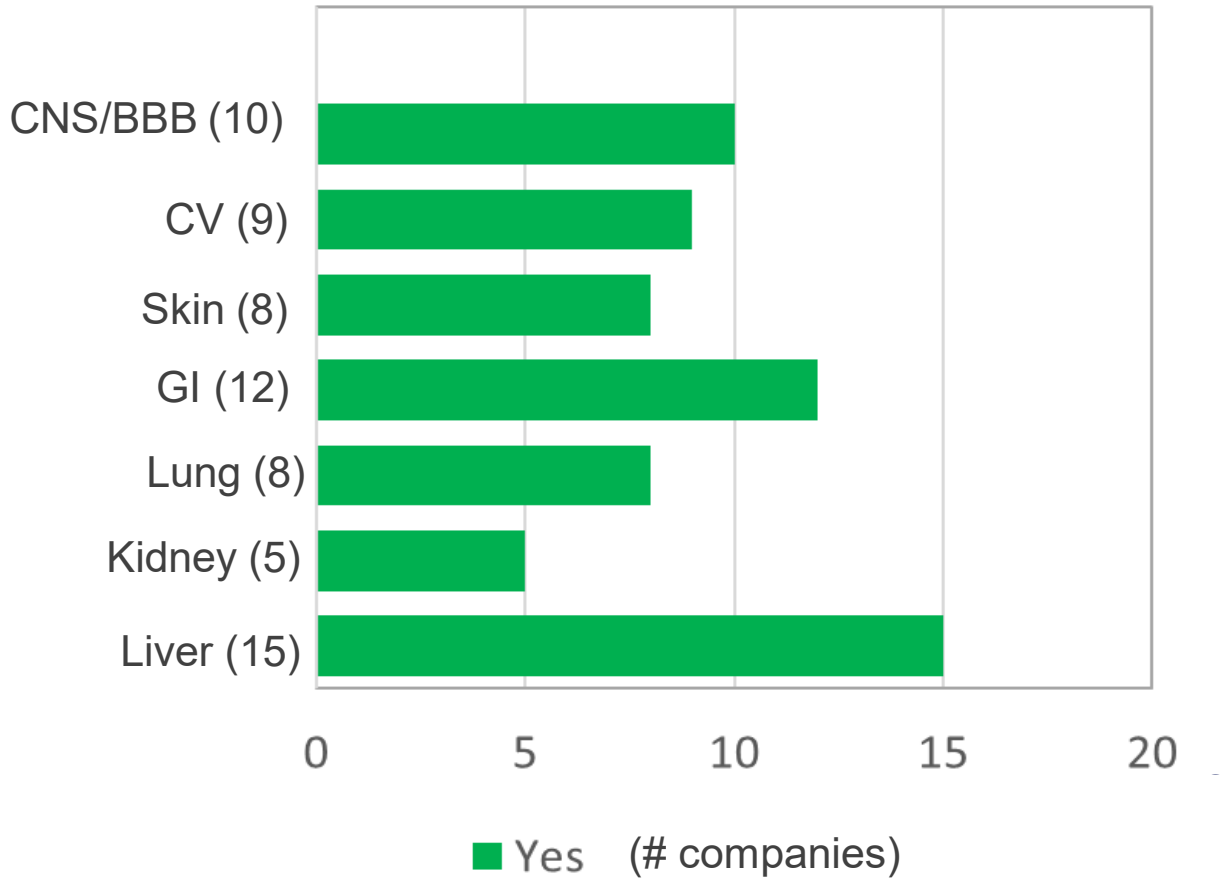


# Landscape Analysis / Gap Assessment Survey Preliminary Results

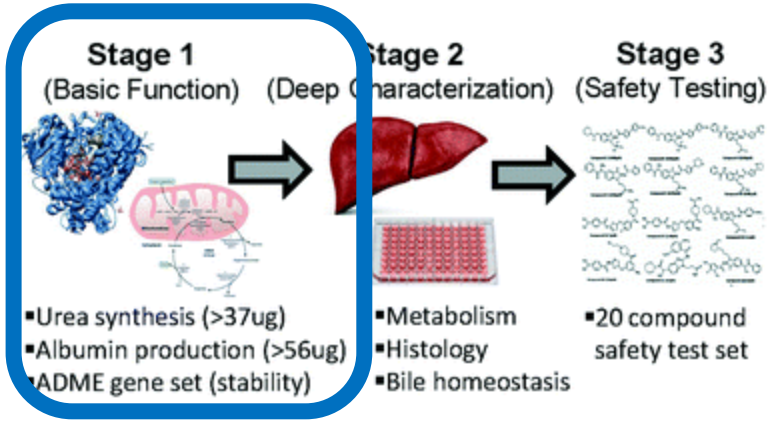
What organ system(s) is your company most interested in modeling with Complex *In Vitro* Models (CIVM)?

| Organ/System | Weighted Ranking |
|---|---|
| Liver | 9.56 |
| Gastrointestinal | 8.06 |
| Kidney | 7.81 |
| CNS/BBB | 7.8 |
| Immune system | 7.6 |
| Lung | 7.09 |
| Cardiovascular | 6.87 |
| Multi-organ systems | 4.73 |
| Skin | 4.58 |
| Reproductive organs | 4.25 |
| Other | 4.1 |

Does your company use complex *in vitro* [organ] models?



# Organ Specific Considerations for Developing, Evaluating and Characterizing MPS Models



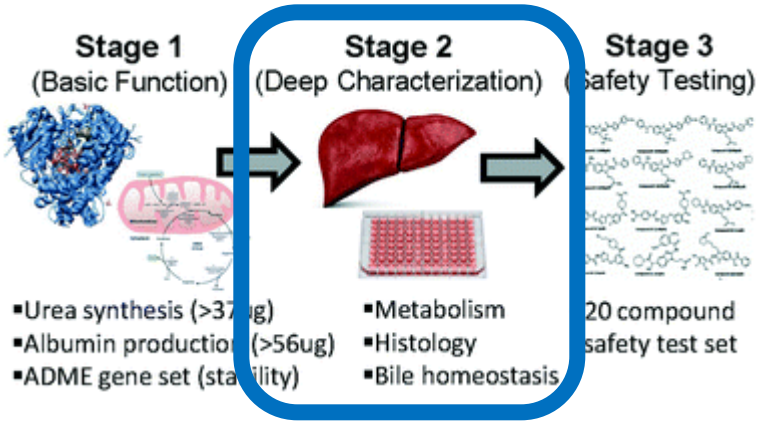

| Measure | Function assessed | Specifications |
|---|---|---|
| Albumin production | Liver transcription, translation, processing, and export function | • >37 μg per day per 1 million hepatocytes<br>• Daily production rates should remain stable across a 14 day time frame<br>◦ Less than a 50% change over a 14 day period with <30% C.V. of mean daily production rates |
| Urea synthesis | Mitochondrial and biochemical synthesis | • >56 μg per day per 1 million hepatocytes<br>• Daily production rates should remain stable across a 14 day time frame<br>◦ Less than a 50% change over a 14 day period with <30% C.V. of mean daily production rates |
| Baseline quantitative gene expression profiling | mRNA expression of ADME genes, stability over time, and levels in comparison to that of a cryopreserved hepatocyte in freshly prepared suspension or human liver sample | • Phase I CYP450 enzymes<br>◦ CYP3A4, CYP2B6, CYP2C9, CYP2C19, CYP1A2, CYP2D6, CYP2C8, CYP2E1<br>• Phase II enzymes<br>◦ UGT1A1, GSTA1<br>• Hepatocyte uptake transporters<br>◦ SLCO1B1, SLCO1B3, SLC22A1<br>• Hepatocyte efflux transporters<br>◦ ABCC2, ABCG2, ABCB1, ABCB11 |

Check for updates

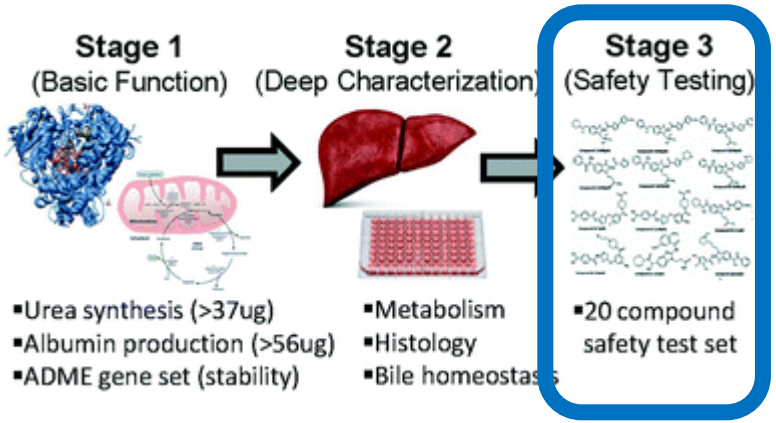
Cite this: *Lab Chip*, 2020, **20**, 215

**Liver microphysiological systems development guidelines for safety risk assessment in the pharmaceutical industry**

Andreas R. Baudy,[a]* Monicah A. Otieno,[b] Philip Hewitt,[c] Jinping Gan,[d] Adrian Roth,[e] Douglas Keller,[f] Radhakrishna Sura,[g] Terry R. Van Vleet[g] and William R. Proctor[h]




| Measure | Function assessed | Specifications |
|---|---|---|
| Alanine aminotransferase (ALT), lactate dehydrogenase (LDH), miR122, cytokines | Indications of cell damage and MPS stability over time | • ≤30% C.V. for mean daily baseline release levels across a 14 day time frame |
| Baseline and induced metabolic enzymes functional activity using a set of standard probe substrates | Liver phase I/II metabolizing enzymes capability (measure of CYP450 enzymatic capacity and induction)<br>Benchmark levels specified for each enzyme compared to fresh hepatocytes and demonstrate <30% CV (as measure of stability of enzymatic activity rates over time) | • **3A4** (midazolam → 1′hydroxymidazolam); show elevated turnover when CYP is induced with rifampicin<br>• **1A2** (phenacetin → APAP); show elevated turnover when CYP is induced with omeprazole<br>• **2B6** (bupropion → hydroxyl bupropion); show elevated turnover when CYP is induced with phenobarbital<br>• **2C9** (diclofenac → diclofenac 2′,3′ oxide or 4-OH diclofenac)<br>• **2D6** (dextromethorphan → dextrorphan)<br>• **UGT1A1** (estradiol → estradiol 3-glucuronide; or 7-hydroxycoumarin → 7-hydroxycoumarin glucuronide)<br>• **GST** (rilpivirine → glutathione conjugate; or dichloronitrobenzene → chloronitrobenzene glutathione) |
| Transporter function and bile acid homeostasis: uptake, metabolism, and export | Measures of daily rates of transporter substrate and bile acid uptake, metabolism, conjugation, and export in media | • Assess transporter functionality using fluorescent probe substrates (*e.g.* cholyl-lysyl-fluorescein for OATP/MRP2/BSEP; tauro-nor-THCA-24-DBD for NTCP/BSEP/MRP2)<br>• Assess bile acid flux using stable label biochemical (*e.g.* labelled GCDCA) with mass spectrometry for bile acid transport |
| Histology of MPS | Allows comparison to that of normal human *in vivo* liver architecture and cellular morphology | • Immunohistochemical analysis of bile canaliculi (BSEP, MRP2, AQP1), Kupffer cells (CD68), and stellate cells (desmin). Electron microscopy of liver sinusoidal endothelial cells to show fenestrations<br>• H&E staining for presence of polygonal, non-rounded, hematoxylin positive, polarized hepatocytes |




| Tool liver toxicant | DILI presentation | Mechanism of toxicity | Appropriate less toxic comparator | DILI presentation | Comparator characteristic |
|---|---|---|---|---|---|
| **Sitaxsentan** | ALT elevations after 2 weeks | Reactive metabolites, mitochondrial toxicity, BSEP inhibition[76–78] | **Ambrisentan** | Minimal ALT elevations[79] | Targets the same receptor as sitaxsentan |
| **Clozapine** | ALT elevations after 1 week | Reactive metabolite[80–82] | **Olanzapine** | No DILI concerns l[83] | Structurally similar to clozapine |
| **Diclofenac** | ALT elevations within 1 month | Reactive metabolites, mitochondrial dysfunction, bile acid dysfunction[84–87] | | | |
| **Zileuton** | ALT elevations after 6 weeks | Reactive metabolite formation[88,89] | | | |
| **Fialuridine** | Liver failure after 12 weeks of dosing | Mitochondrial toxicity as primary event causing lactic acidosis, microvesicular steatosis[90] | **FIRU [1-(2′-fluoro-2′-deoxy-D-ribofuranosyl)-5-iodouracil]** | No DILI concerns | Stereoisomer of fialuridine. Only *in vitro*/animal data available |
| **Tolcapone** | ALT elevations, acute liver failure | Reactive metabolite, mitochondrial toxicant, BSEP inhibition[45,91] | **Entacapone** | Low DILI concern[92] | Similar BSEP profile, but less mitochondrial toxicity |
| **Asunaprevir** | ALT elevations after 2 weeks[93] | Alterations in bile acids | | | |
| **Troglitazone** | ALT, bilirubin elevations after 18 weeks | Reactive metabolites, BSEP inhibition[94–96] | **Pioglitazone** | Low DILI concern[97] | Low clinical dose |
| **Telithromycin** | ALT elevations after 1 day | Bile acid alterations[98,99] | | | |
| **Trovafloxacin** | Acute liver failure | Immune mediated[100] | **Levofloxacin** | No/low DILI concern[101] | Same structural class as trovafloxacin |
| **Pemoline** | Liver failure | Immune mediated[102] | | | |
| **Mipomersen** | Oligonucleotide, ALT elevations and hepatic steatosis | Lipid alterations[103] | | | |
| **Nefazodone** | Liver failure | Reactive metabolites, BSEP inhibitor, mitochondrial tox[54,104] | **Buspirone (or Trazodone)** | No/low DILI concern[105] | Less BSEP inhibition and weak to no mitochondrial toxicity |

# Examples of General Considerations for Developing, Evaluating and Characterizing MPS Models

| Functional characteristic | Present/ Retained | Assessed (Y/N) | Assessment performed | References |
|---|---|---|---|---|
| Permeability | | | | |
| Transmigration | | | | |
| Cell differentiation | | | | |
| Cell proliferation | | | | |
| Cell functional maturation | | | | |
| Cell functional stability | | | | |
| Cell regeneration/recovery response | | | | |
| Expression of receptors | | | | |

# Examples of General Considerations for Developing, Evaluating and Characterizing MPS Models

| Structural characteristics | Present/ Retained | Assessed (Y/N) | Assessment performed | References |
|---|---|---|---|---|
| Cell ratio | | | | |
| Cell orientation and polarity | | | | |
| Physicochemical properties of extracellular matrix material (including charge and chemical composition) | | | | |
| Cell–cell interactions | | | | |
| Tissue microenvironment | | | | |
| Tissue – tissue interaction | | | | |
| Structure-function relationship | | | | |

| System components | Present | Material | Nonspecific binding / diffusion (Y/N) | Nonspecific binding / diffusion (value) | References |
|---|---|---|---|---|---|
| Scaffold / matrix | | | | | |
| Membrane | | | | | |
| Membrane coating | | | | | |

| Cellular components | Type (iPS, ES cells, primary, etc.) | Source | Viability duration | Functionality retained | Assessment type | References |
|---|---|---|---|---|---|---|
| Neural | | | | | | |
| Vascular | | | | | | |
| Immune | | | | | | |
| Other | | | | | | |

| | Available / Retained / Capability | Assessed (Y/N) | Assessment performed | References |
|---|---|---|---|---|
| Functional readout(s) | | | | |
| Cellular phenotype(s) | | | | |
| Exposure; acute | | | | |
| Exposure; chronic | | | | |
| Throughput | | | | |
| Mechanical stimuli; flow | | | | |
| Mechanical stimuli; stretch | | | | |
| Live imaging | | | | |

# Innovators Influencing Engagement via Information Sharing

**Duration**

- Set up time including cells
- Viability
- Activity/metabolic functionality

**System**

- Capacity
- Maintenance level
- Throughput
- Space requirements
- Equipment requirements
- Material properties (compound binding)
- Level of training/expertise required

**Abilities**

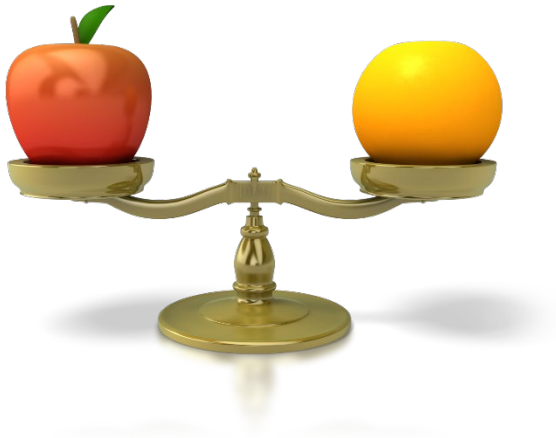
- Sampling
- Frequency (some systems do not allow for daily sampling)
- Type (liquid, histology)
- Imaging
- *In situ*

**Testing parameters**

- Cell sourcing including commercial *versus* non-commercial
- Media sourcing including commercial *versus* non-commercial
- Reproducibility level
- Comparisons
- 2D systems
- *In vivo* models
- Baseline function assays
- Toxicity assays
- Appropriate positive/negative controls

**Restrictions**

- In house only
- Limited cell types



**Business model**

- For customer use
- Contractual (in house only)

**Testing parameters**

- Cell sourcing including commercial *versus* non-commercial

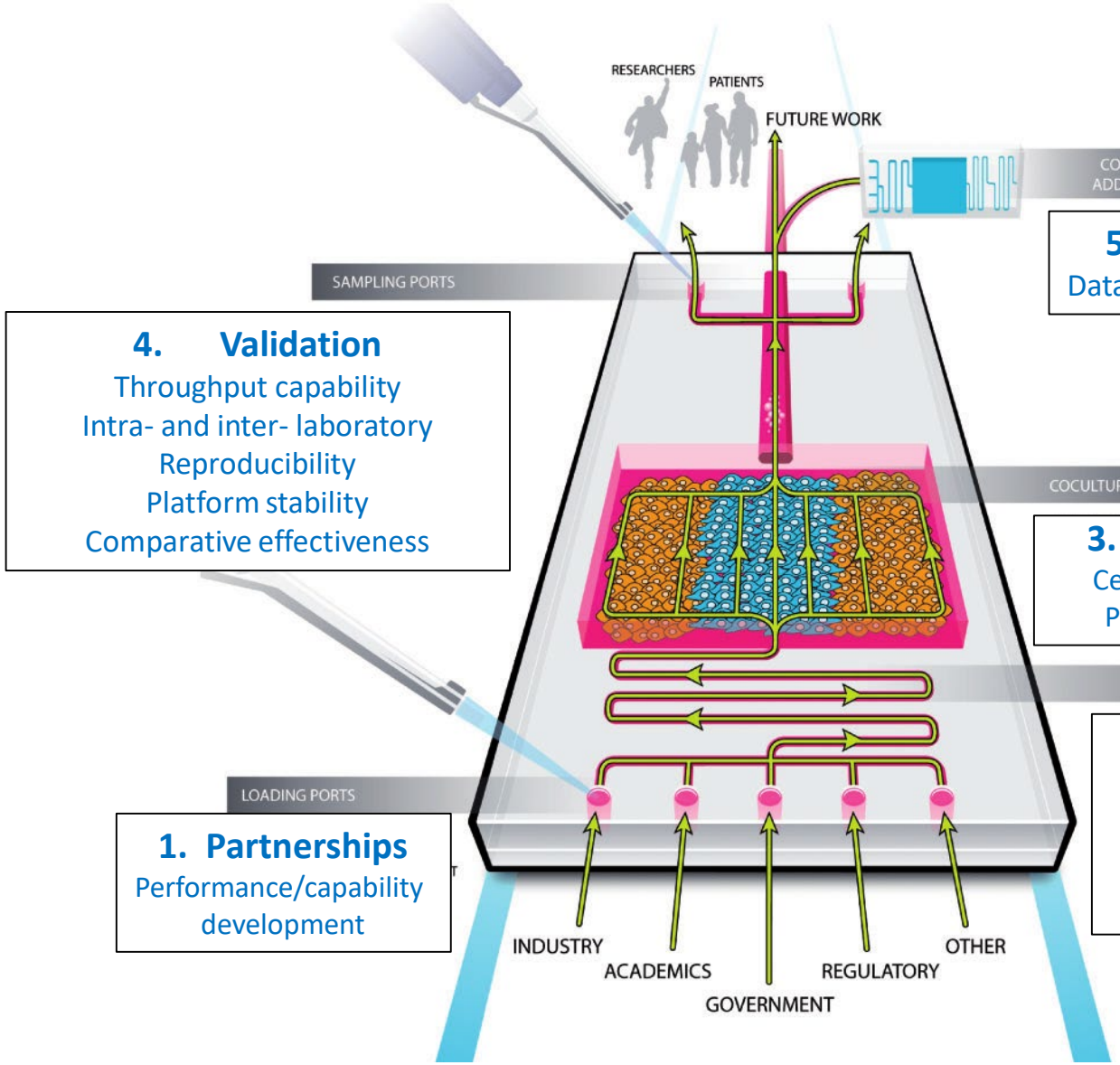
# Conclusions

- Knowledge of contemporary or standard *in vitro* models and practices
- Use compounds/molecules that have high value for industry
- Understand
  - Context of use
  - Limitations
- Practical and scalable
- Need to provide additional value

# Advancing MPS Technologies Through Collaboration

5–10+ yrs

Current approach




**1. Partnerships**
Performance/capability development

**2. Defining Context of Use**
Mechanistic assays
Safety screening
DMPK/Disposition
Efficacy

**3. Characterization**
Cell/Tissue Composition
Physiologic Function…

**4. Validation**
Throughput capability
Intra- and inter- laboratory Reproducibility
Platform stability
Comparative effectiveness

**5. Commercialization**
Data and decision management

2–5 yrs

Pharma leadership

Adopted from L. Ewart, K. Fabre, A. Chakilam, Y. Dragan, D. B. Duignan, J. Eswaraka, J. Gan, P. Guzzie-Peck, M. Otieno, C. G. Jeong, D. A. Keller, S. M. de Morais, J. A. Phillips, W. Proctor, R. Sura, T. Van Vleet, D. Watson, Y. Will, D. Tagle and B. Berridge, **Navigating tissue chips from development to dissemination: A pharmaceutical industry perspective.** *Exp. Biol. Med.*, 2017, **242**, 1579–1585

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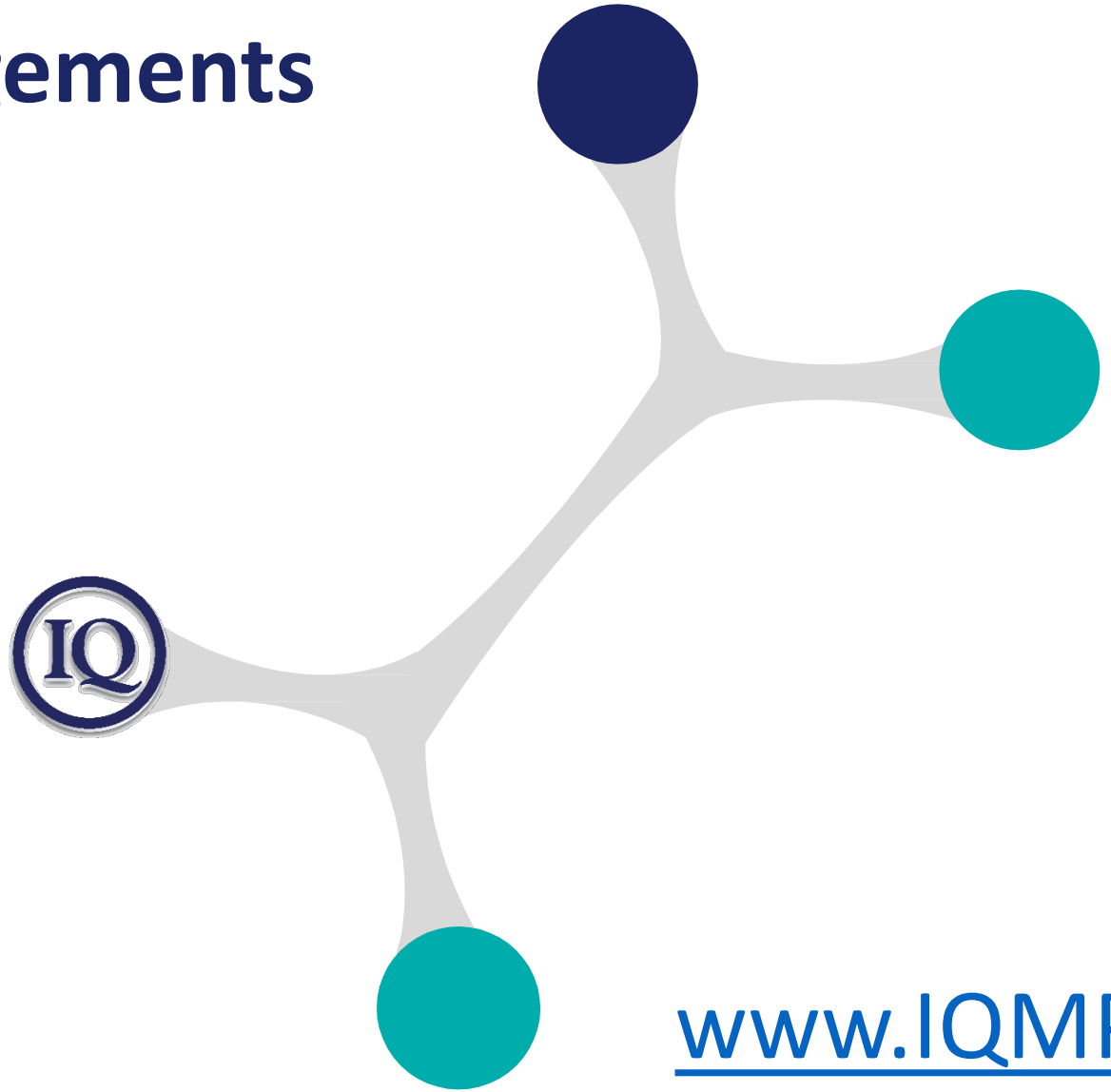

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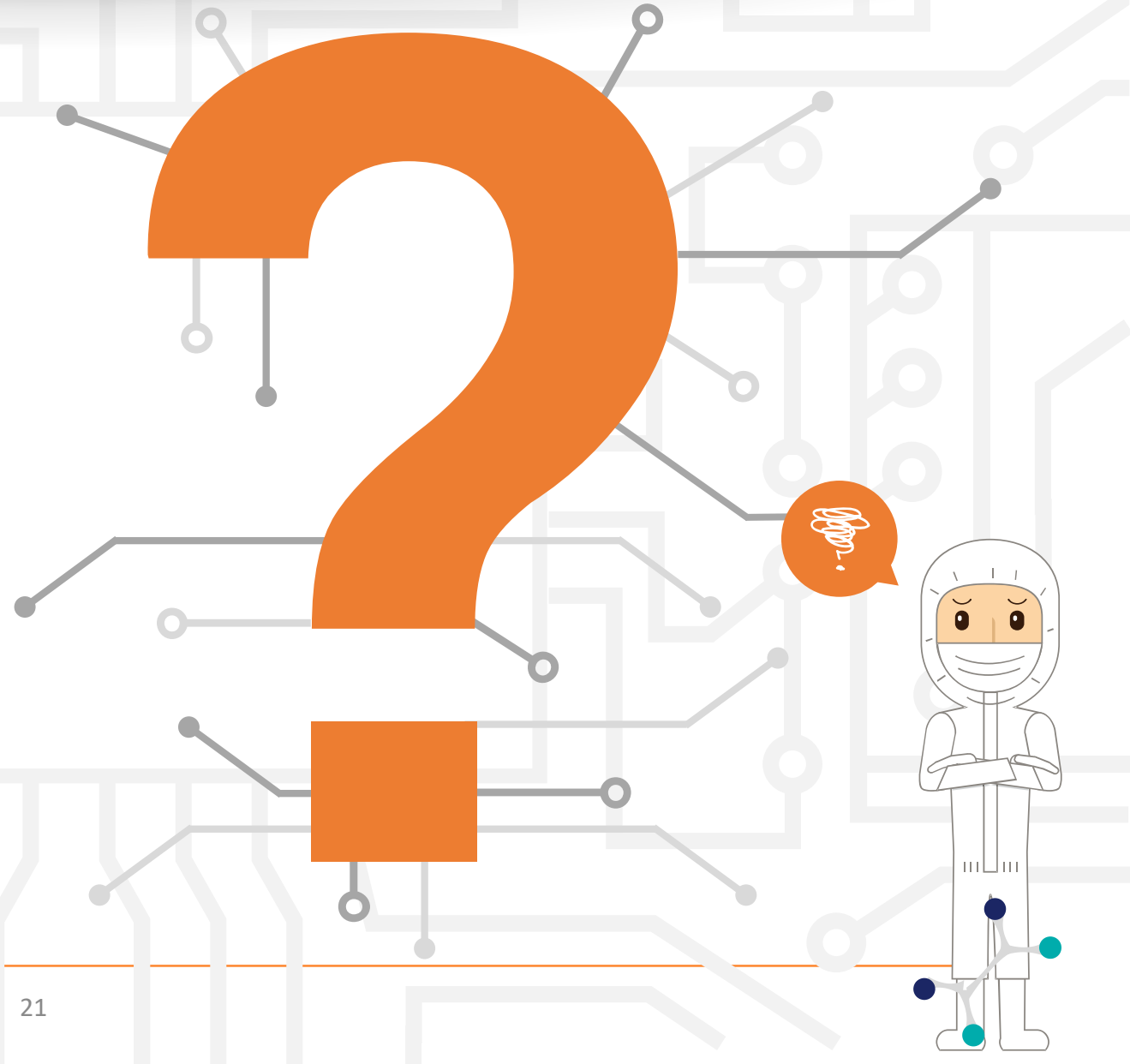
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