

# A primary jejunum and primary hepatocyte multi-organ MPS: a promising tool for more predictive studies of human drug ADME and oral bioavailability

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## Abstract

Traditional immortalized intestinal cell lines and suspension hepatocytes have absent or low levels of metabolic enzyme expression, and thus fail to predict first pass human metabolism and oral bioavailability.

Efforts to improve the *in vitro* to *in vivo* translation of drug efficacy and safety data has led to the emergence of more human relevant microphysiological systems (MPS) that consist of multiple, fluidically linked organs. Here we describe a new MPS that links the jejunum (RepliGut<sup>®</sup> planar - jejunum) and liver using the PhysioMimix<sup>®</sup> Multi-organ System, with both cell types being of primary human origin.

To demonstrate improved predictive capacity, we investigated two drugs where current models fail to adequately predict human ADME behaviour. Temocapril, which is a prodrug and is designed to be resistant to intestinal hydrolysis<sup>2</sup> and midazolam, which is known to undergo intestinal clearance<sup>3</sup>.

## Objectives

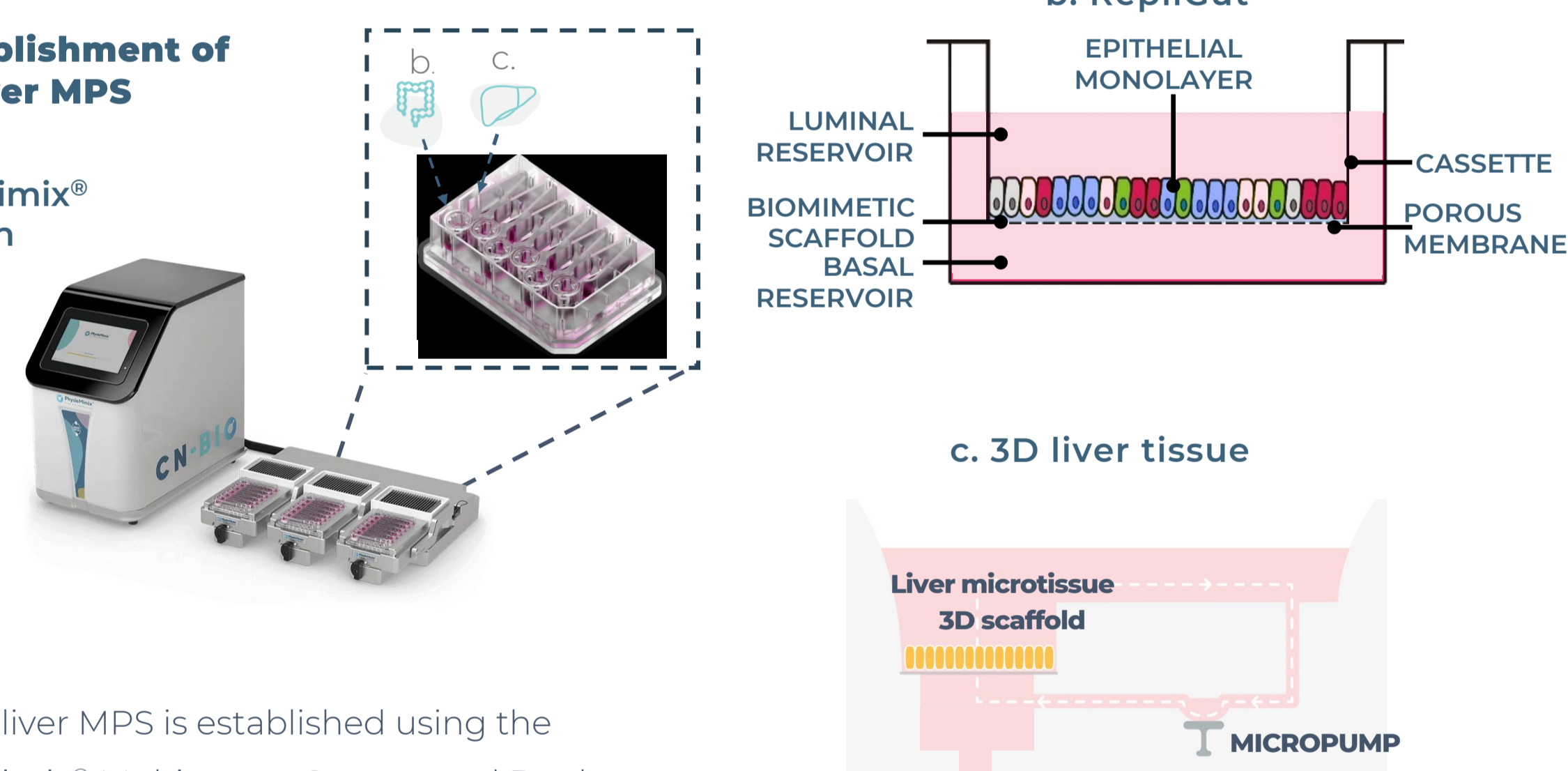
The objectives of this study are:

- Develop a gut-liver MPS made up of entirely primary human cells
- Demonstrate the functionality of liver and intestinal tissues
- Deliver a protocol to conduct drug ADME and bioavailability studies
- Demonstrate improved predictive capacity with two drugs where current models fail to predict human ADME behaviour

## Methods

**Fig 1. Establishment of the gut-liver MPS**

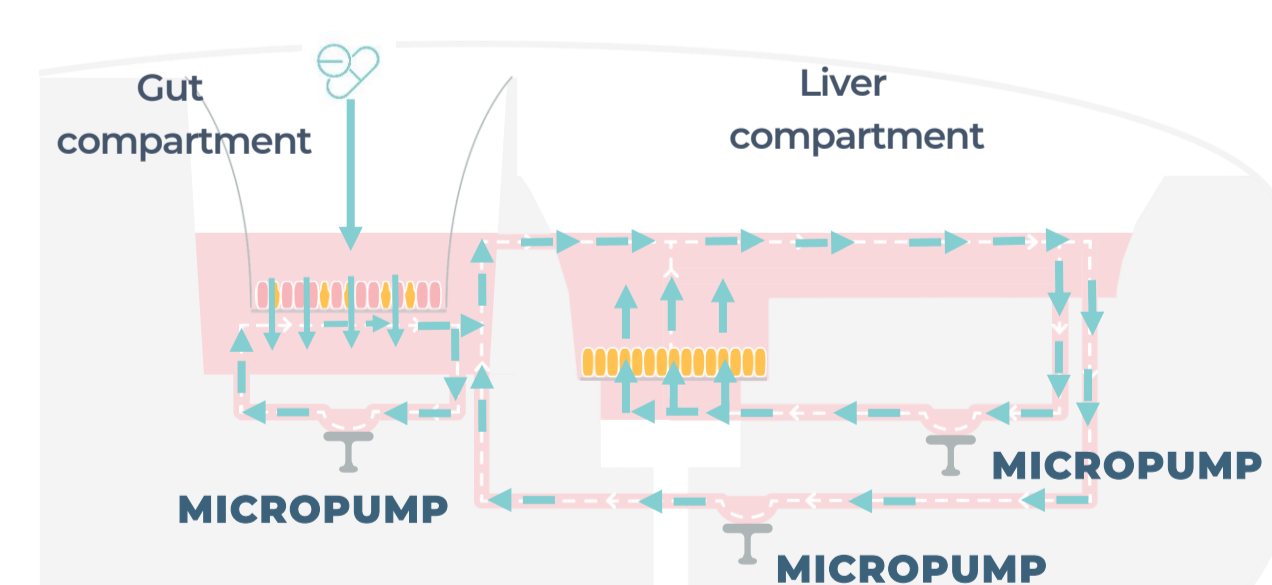
a. PhysioMimix<sup>®</sup> Multi-organ System



- The gut-liver MPS is established using the PhysioMimix<sup>®</sup> Multi-organ System and Dual-organ plate (Figure 1 a).
- Liver microtissues are formed by seeding primary human hepatocytes (PHH) on a 3D collagen coated scaffold (Figure 1 c).
- For the intestinal barrier, jejunum crypt epithelial stem cells were expanded and differentiated on a biomimetic scaffold, in static conditions (RepliGut<sup>®</sup>, Figure 1 b).
- 4 days post PHH seeding, RepliGut<sup>®</sup> is added into the gut compartments of Dual-organ plates and fluidically connected to its respective liver compartment. (Figure 2). ADME studies begin.

**Fig 2. ADME Study**

Drug dosed (temocapril= 100 µM, midazolam= 50 µM) in the gut apical compartment. After transport through the barrier, the drug is mixed into the gut basolateral and liver compartments



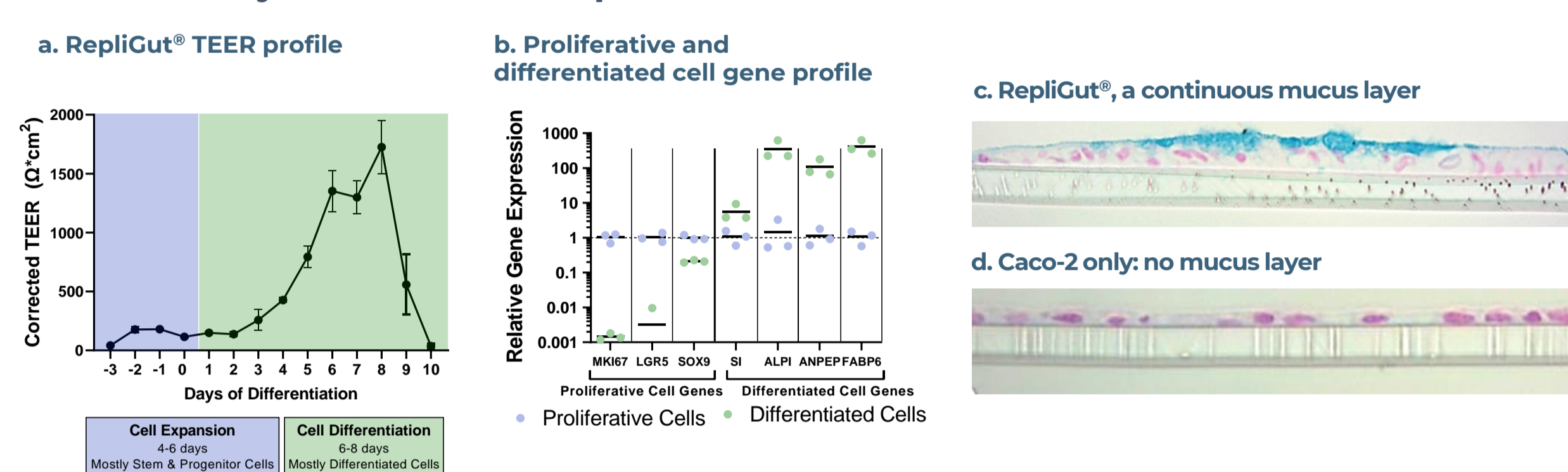
## Conclusion

The gut-liver MPS recapitulates the physiological conditions of oral drug dosing in the human (Figure 3-5). Made up entirely of primary human cells, this model enables for more predictive ADME and bioavailability studies compared to standard *in vitro* models (Figure 7).

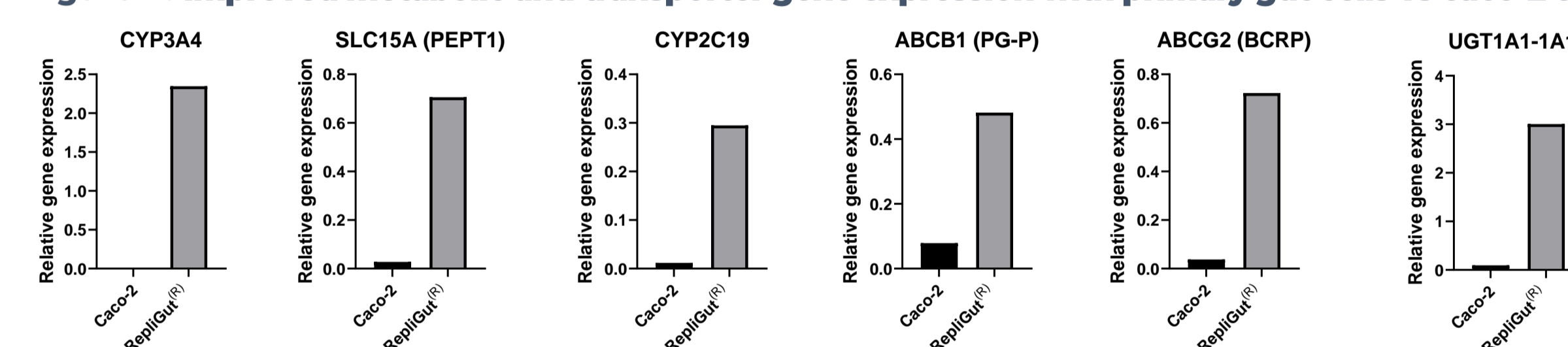
This human relevant gut-liver model offers a vast improvement in the methods used to study the pharmacokinetics of prodrugs that undergo CES metabolism, such as temocapril (Figure 6).

## Results

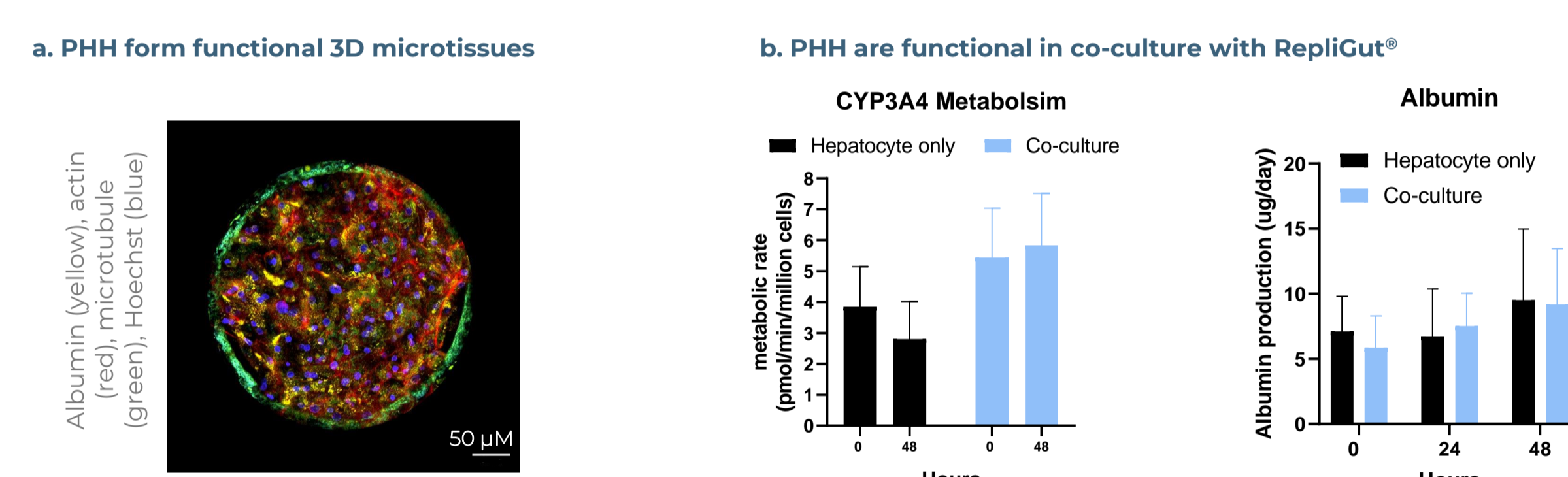
**Figure 3. RepliGut<sup>®</sup> exhibits distinct proliferative and differentiated cell populations and a continuous layer of mucus on its apical surface**



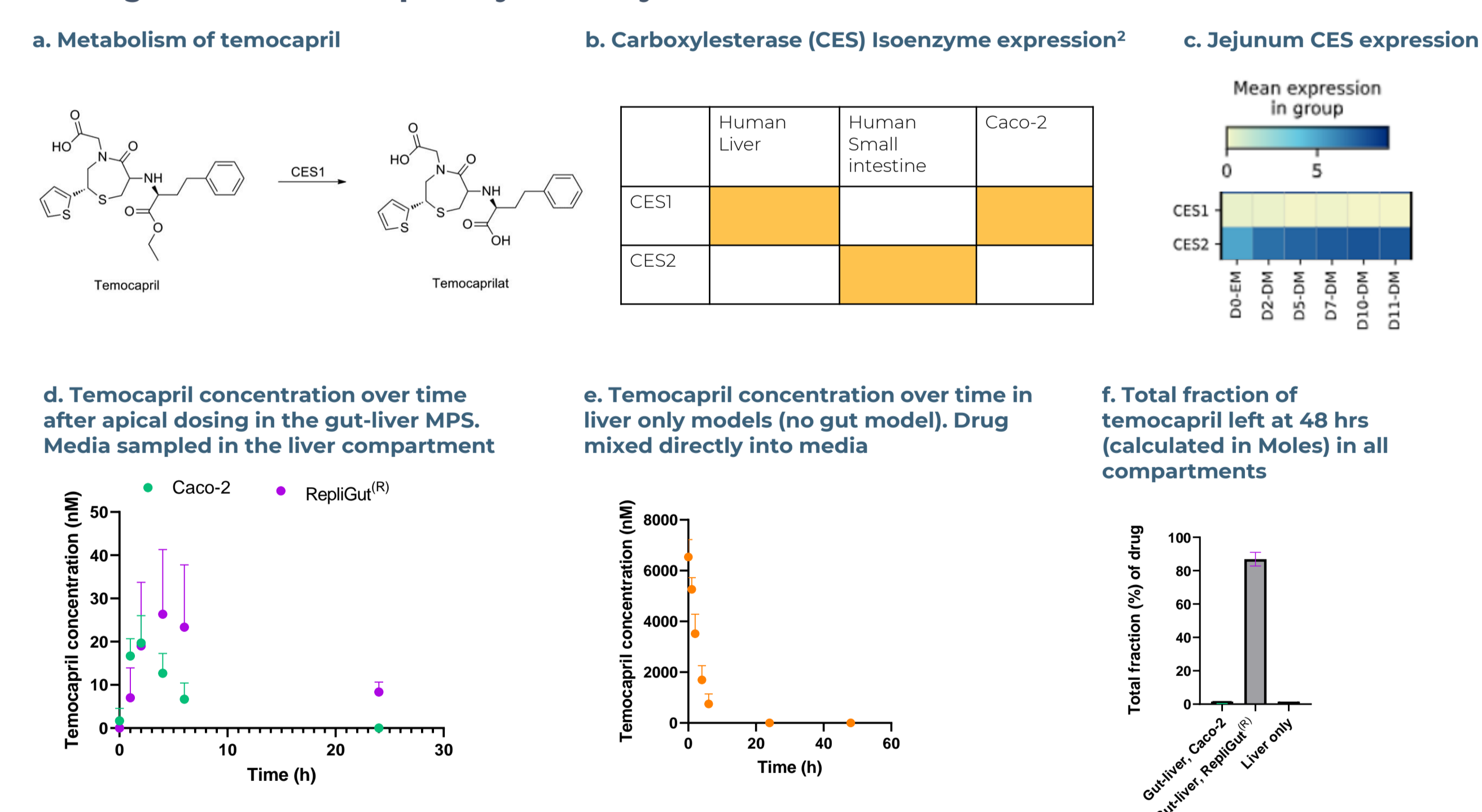
**Figure 4. Improved metabolic and transporter gene expression with primary gut cells vs caco-2 cell line**



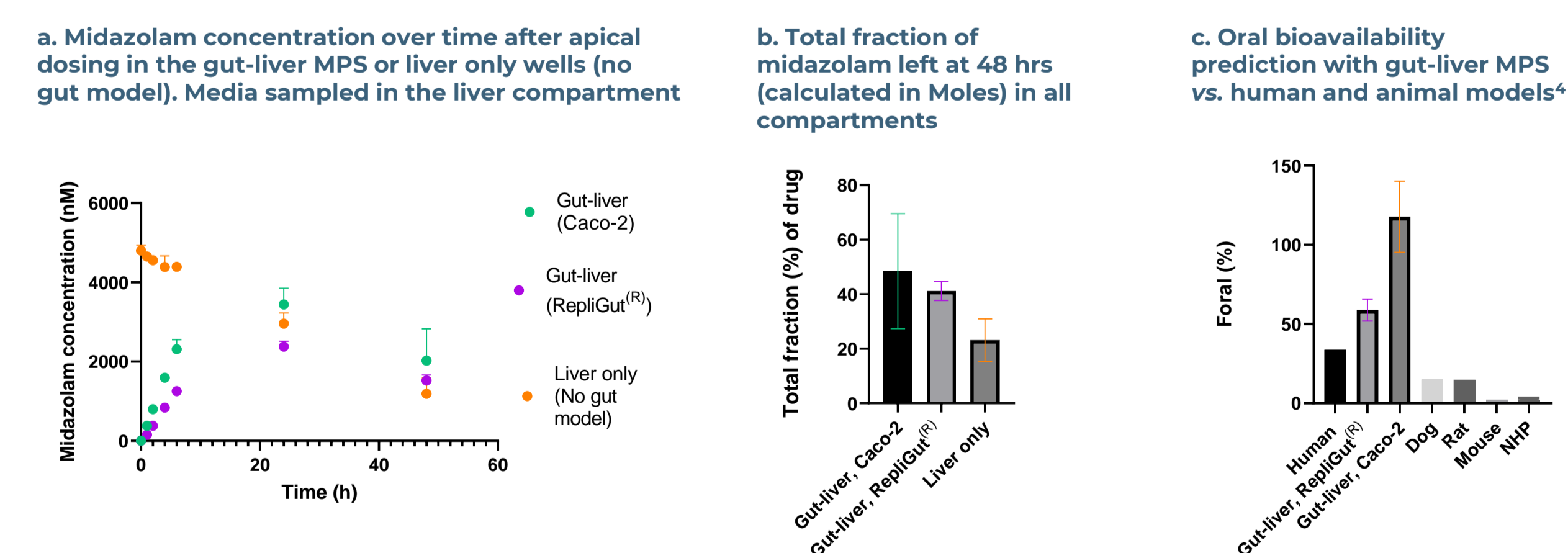
**Figure 5. 3D liver tissues are functional and metabolically active in both liver only and gut-liver co-culture.**



**Figure 6. Clearance of temocapril correlates with human isoenzyme expression in the gut-liver MPS with primary cells only**



**Figure 7. Improved bioavailability prediction of midazolam in the gut-liver MPS with primary cells**



## References

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