

# Extending the choice of ULA plates for primary 3D human liver spheroid formation and experimentation with the Akura™ Spheroid Microplates



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## Introduction

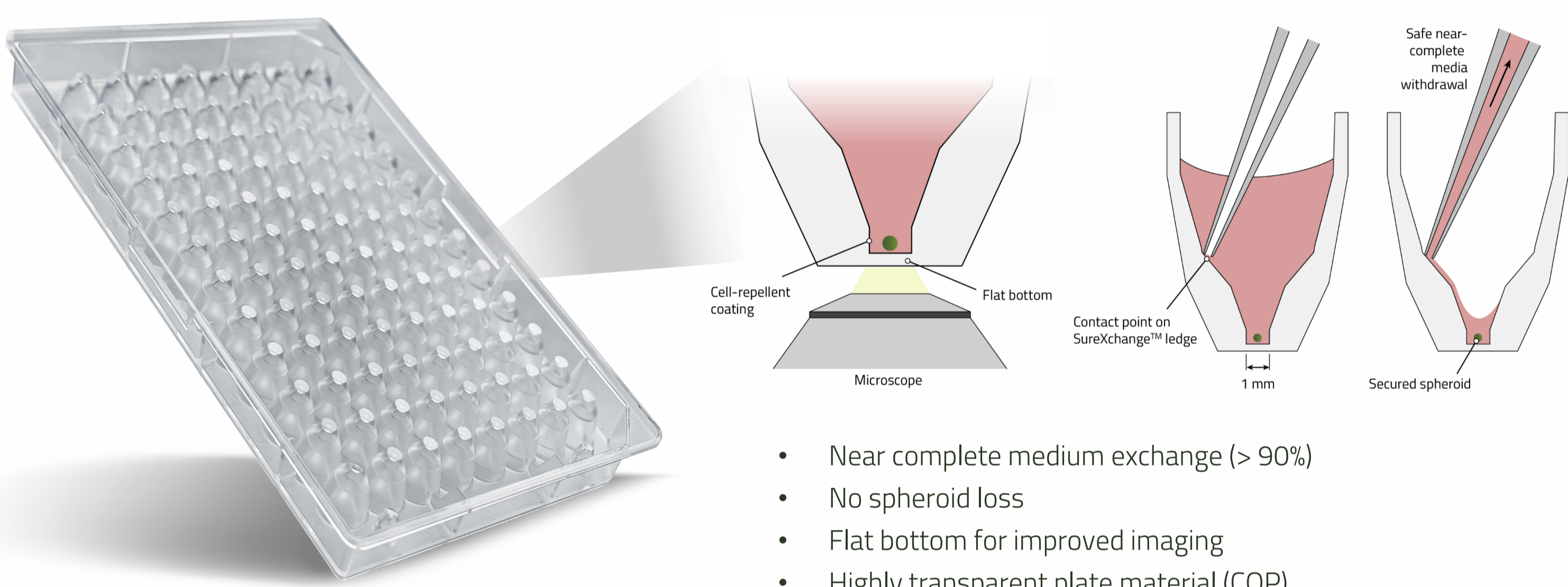
The plate matters - Scale up and routine application of 3D cell cultures, such as spheroids, require selecting the right and matching hardware. Plate design, features, surface property and production quality largely determine the tissue quality, reproducibility and organ-specific function of the spheroid. Further, the plates can provide different readout accessibility for spheroid analysis. In a comprehensive study entitled "The choice of ultra-low attachment plates impacts primary human and primary canine hepatocyte spheroid formation, phenotypes, and function" [1] Xing *et al.* compared several commercially available 96- and 384-well plates for primary liver spheroid aggregation, culturing and analysis. The study addresses aggregation efficiency and quality, their compatibility for imaging, spheroid viability and robustness over time as well as the preservation of liver-specific function and response to known toxic compounds.

Here, we provide comparative data obtained from liver spheroid production runs and their characterization in InSphero's Akura™ 96 and 384 Spheroid Microplates. The aim is to further expand this study and broadening the choice for spheroid users to select the right plate for their application.

[1] Xing, C., Kemas, A., Mickols, E., Klein, K., Artursson, P., & Lauschke, V.M. (2024). The choice of ultra-low attachment plates impacts primary human and primary canine hepatocyte spheroid formation, phenotypes, and function. *Biotechnology Journal*, 19, e2300587. <https://doi.org/10.1002/biot.202300587>

## Akura™ 96 Microplate

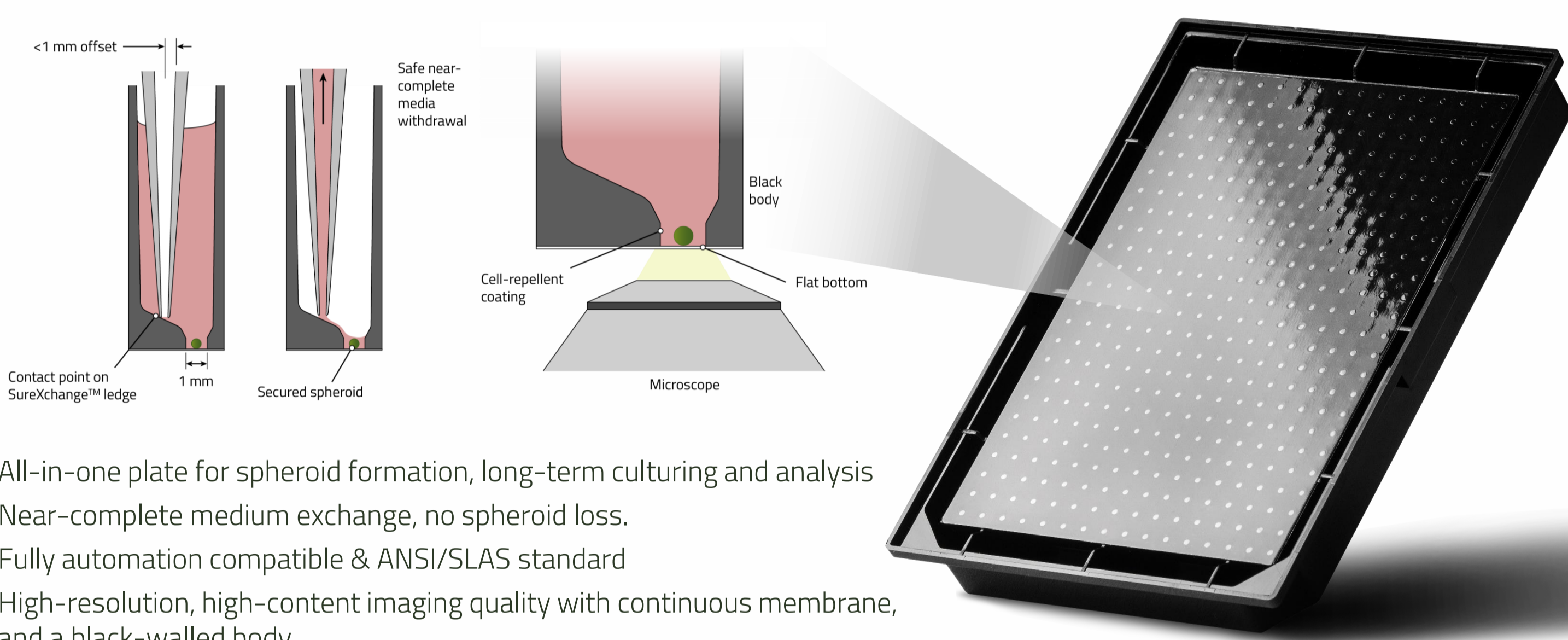
Unique well design – Secure handling – Automation compatible.



- Near complete medium exchange (> 90%)
- No spheroid loss
- Flat bottom for improved imaging
- Highly transparent plate material (COP)

## Akura™ 384 Microplate

High throughput screening with robust automation.



- All-in-one plate for spheroid formation, long-term culturing and analysis
- Near-complete medium exchange, no spheroid loss.
- Fully automation compatible & ANSI/SLAS standard
- High-resolution, high-content imaging quality with continuous membrane, and a black-walled body

## Liver Spheroid Aggregation

High uniformity and reproducibility across plates and production lots.

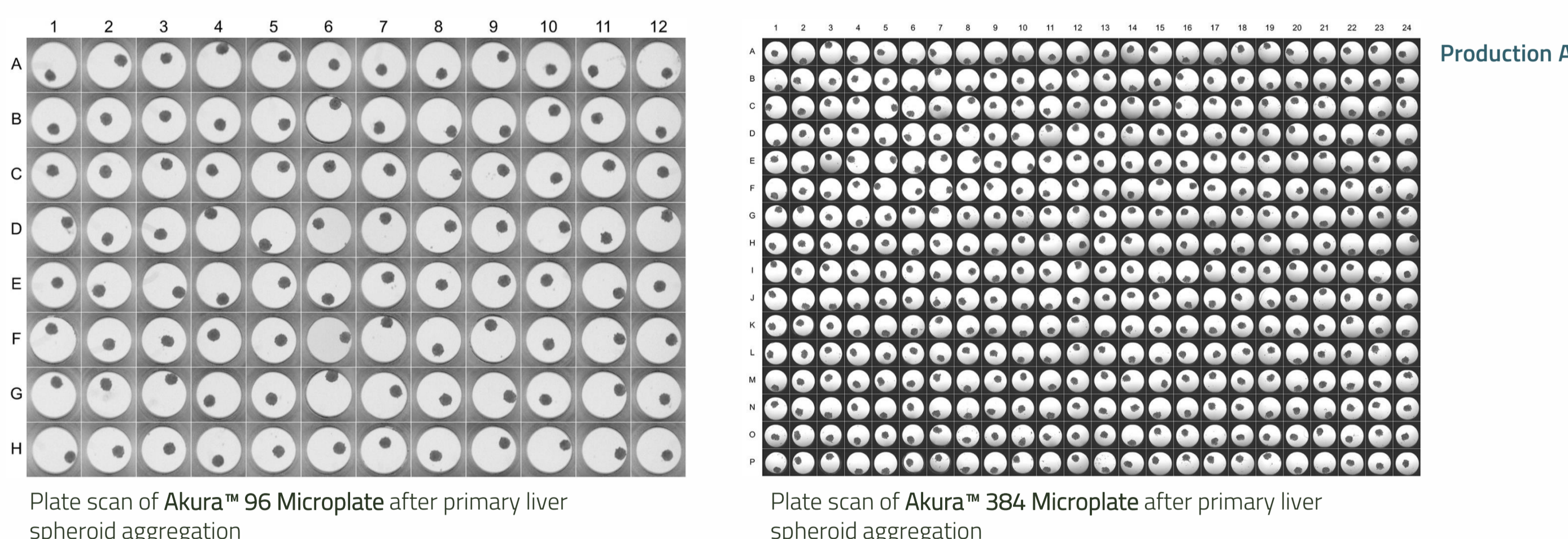
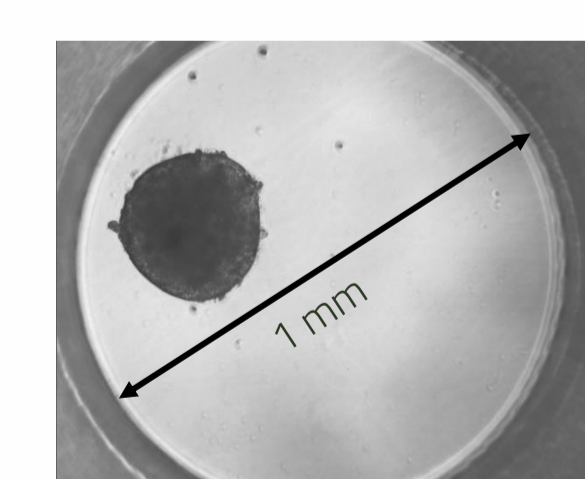


Plate scan of Akura™ 96 Microplate after primary liver spheroid aggregation

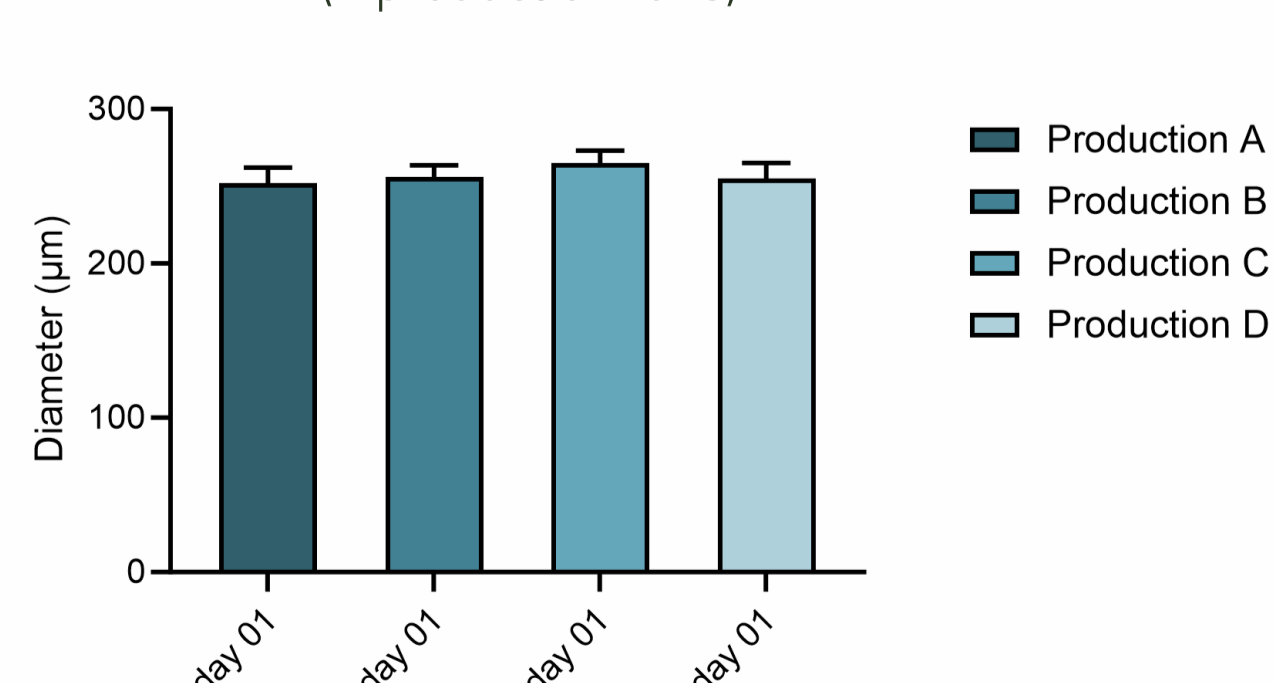
Plate scan of Akura™ 384 Microplate after primary liver spheroid aggregation



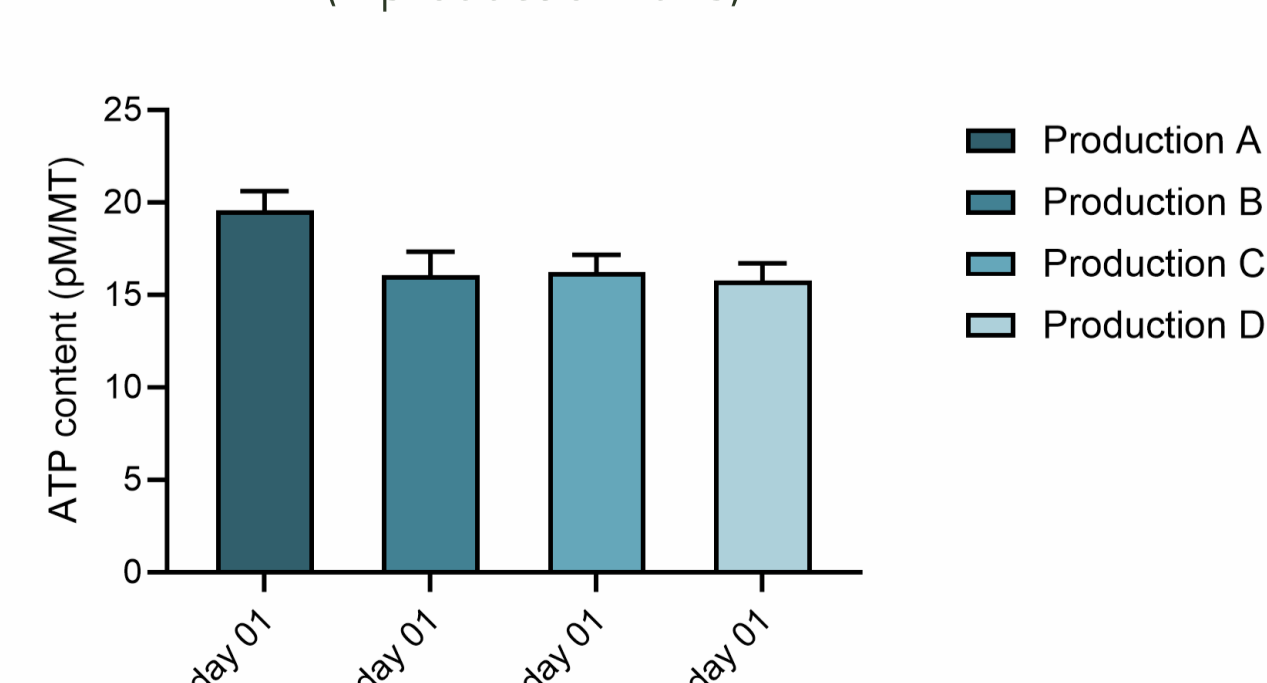
• 1-mm observation chamber simplifies spheroid localization, and ROI identification

- Rapid aggregation within 4-5 days
- Compact spheroids morphology without satellites

Liver spheroid diameter at Day 1 (4 production runs)



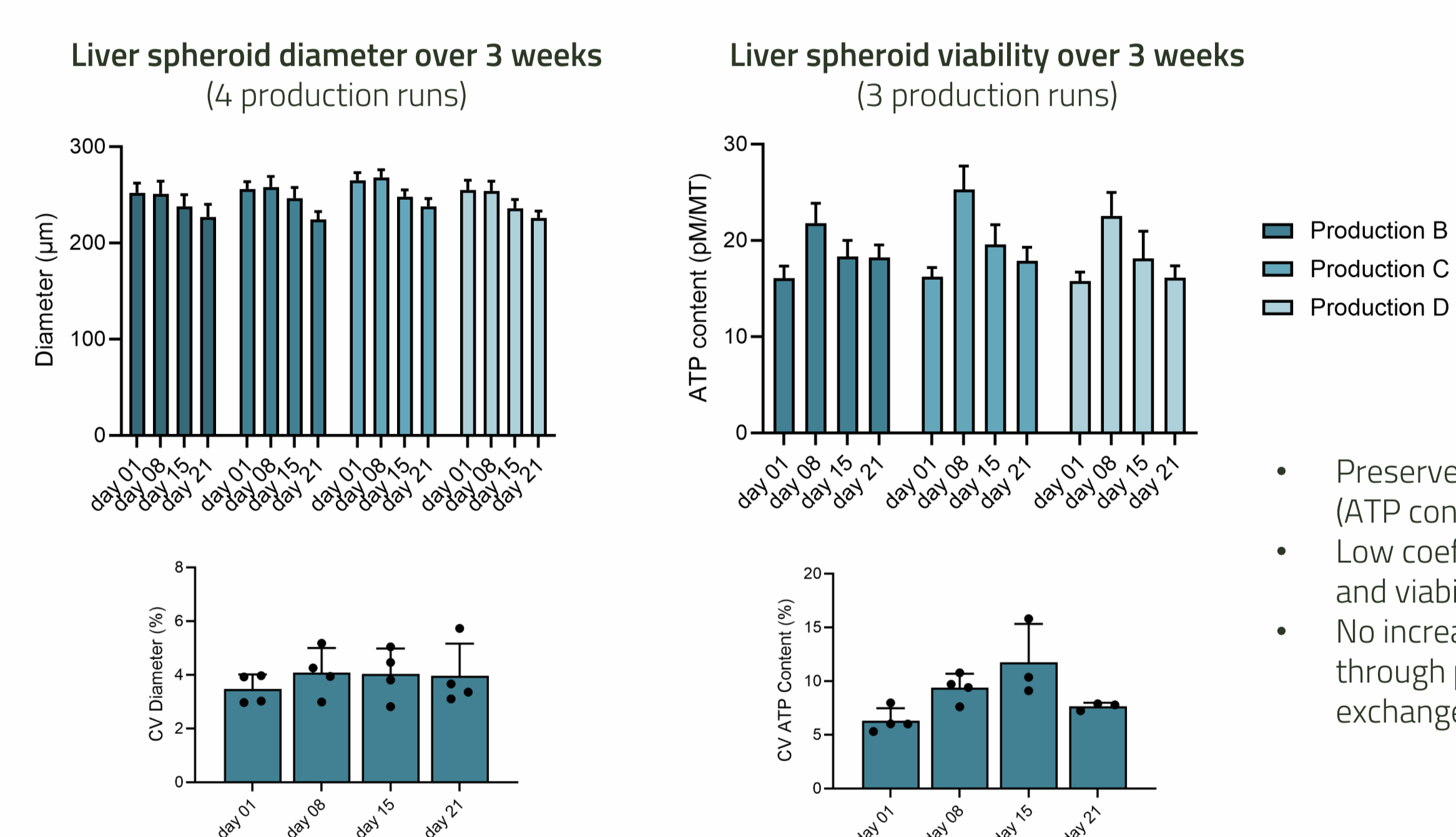
Liver spheroid viability at Day 1 (4 production runs)



- Low well-to-well variability of spheroid sizes
- Consistent size of spheroids across production runs
- Low well-to-well variability of spheroid viability
- Consistent ATP content of spheroids across production runs

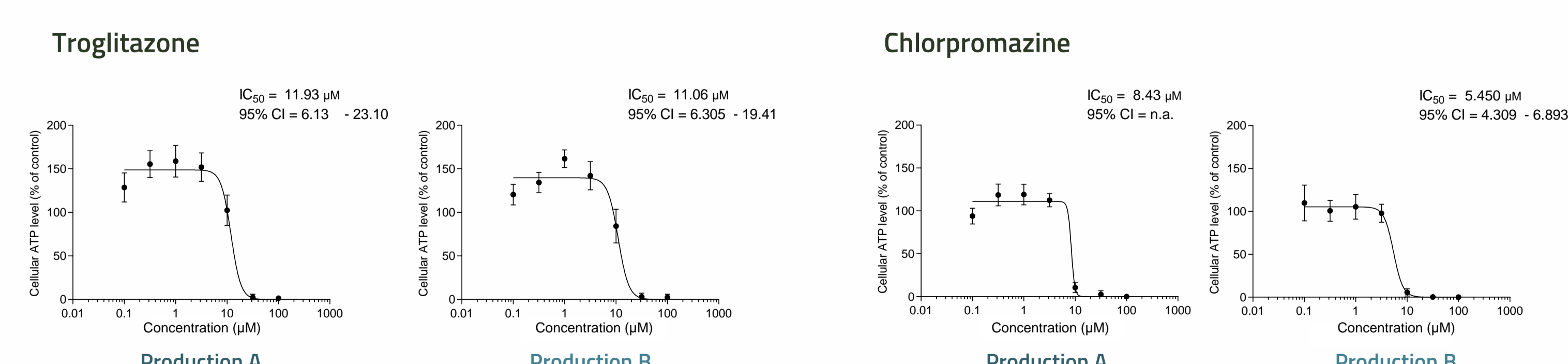
## Liver Spheroid Stability

Stable spheroid size and viability over time with low variation.



- Preserved spheroid size and viability (ATP content) over 3 weeks
- Low coefficient of variation in size and viability across production runs
- No increase in variability over time through precise and robust medium exchange

## Dose Response to Reference Compounds



- Consistent IC<sub>50</sub> values of reference compounds for drug-induced liver injury across production runs