

MHCP-I-3D Mouse Hepatocytes Cryopreserved Plateable for Induction assays and for 3D culture
Cell Specification – Certificate of Analysis (CoA)

Lot MH240618

Batch Release: December 16, 2024

Donor data

Species: Mouse (*Mus musculus*)
Strain: Swiss CD-1 // RjOrl:SWISS

Gender: male
Age: approx. 4 months

Cryopreservation and Thawing

Cryopreservation:

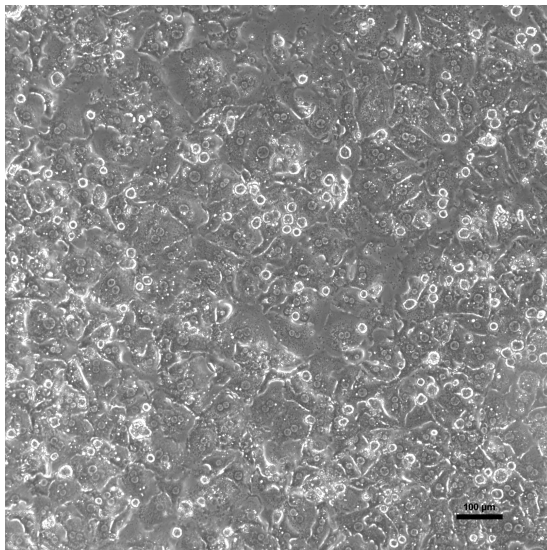
Date: Jun 18, 2024
Amount per vial: 6.75×10^6 cells

Thawing: n=1

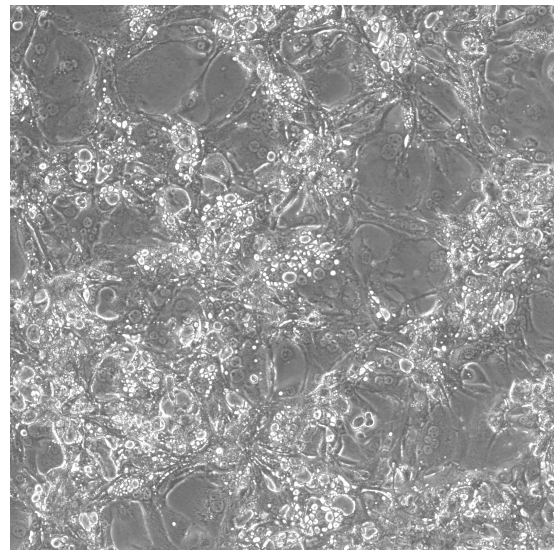
Post-thaw viability: 76.3 %
Post-thaw yield per vial: 2.06×10^6 cells
Recovery: 30.5 %

2D culture

Phase contrast on day 1 after thawing
(24well plate)



Phase contrast on day 3 after thawing
(24well plate)



Recommended seeding density on collagen-coated plates:

24well plate – 160,000 cells/well // 96well plate – 20,000 cells/well

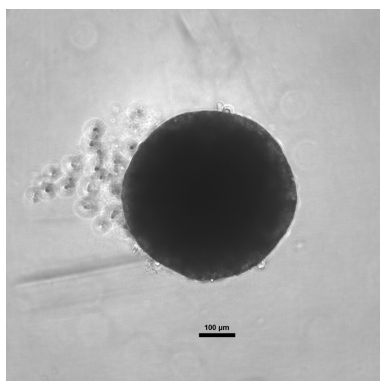
Culture in Human Hepatocyte Maintenance Medium (HHMM).

CYP P450 activity in 2D culture after thawing:	pmol/ (mg × min)	X-fold induction
Ethoxyresorufin-O-deethylation:	24well: $10.5 \pm 0,3$	3.1
Induction with 25 µM β-Naphthoflavone	96well: 15.4 ± 3.1	3.6

3D culture

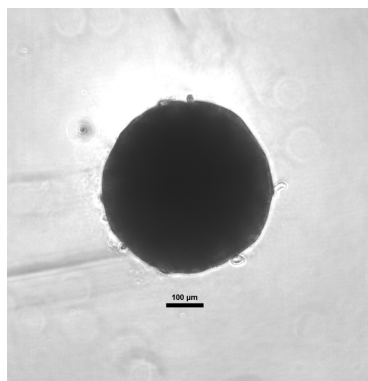
Cells seeded in 96well ULA round bottom plates (FaCellitate), 2,500 cells/well

day 14



scale bar 100 µm

day 18



scale bar 100 µm

Suspension culture

Viability test on orbital shaker (Eppendorf Thermomixer C, 1000 rpm at 37 °C with 0.5×10^6 cells in 0.5 ml HPM-Cryo):

Time (h)	0	1	2	3	4	5
Viability (%)	76.3	58.8	61.6	50.5	49.7	47.6

Note: Yield, viability, recovery and activity assays were performed at PRIMACYT using PRIMACYT's manual for thawing, plating and culture of primary cryopreserved hepatocytes.

Store at -150 °C or in the vapour phase of LN₂

This product should be considered as potential biohazard. Only intended for *in vitro* use.

Detailed animal information and husbandry conditions	
Species	Mouse (<i>Mus musculus</i>)
Strain	Swiss CD-1 (RjOrl:SWISS)
Vendor	Janvier Labs Saint Berthevin Cedex, France
Food	Altromin Maintenance diet for rats/mice (ad libitum), Altromin, Lage, FRG
Water	ad libitum
Light/Dark cycle	regulated day/night cycle using artificial and natural light, 10-14 hours light daily
Temperature	20-24 °C
Humidity	40-70 %
Bedding	Hugro hemp bedding, Saerbeck, FRG
Cage	Tecniplast Eurostandard Type III and Type IV incl. behavioural enrichments for the animals
<p>Animals were housed under veterinary control and allowed to acclimate ≥ 7 days before use. Animal housing permits according to §11 Abs. 1 TSchG, dated March 22, 2022 under supervision of Veterinary Office of Landkreis Ludwigslust/Parchim, FRG. Hepatocytes were obtained from a non-infectious, non-contagious, healthy animal. The animal does not originate from a facility conducting work or research with animal pathogens.</p>	

Issued by: A. Alkufairi	Verified by: T. Krimmling
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