

**MH-MHCP Mouse Hepatocytes Cryopreserved Plateable for Induction assays
Cell Specification – Certificate of Analysis (CoA)**

Lot MH240111-2

Batch Release: April 23, 2024

Donor data

Species: Mouse (*Mus musculus*)
Strain: Black 6 (C57BL6JR)

Gender: male
Age: approx. 6 months

Cryopreservation and Thawing

Cryopreservation:

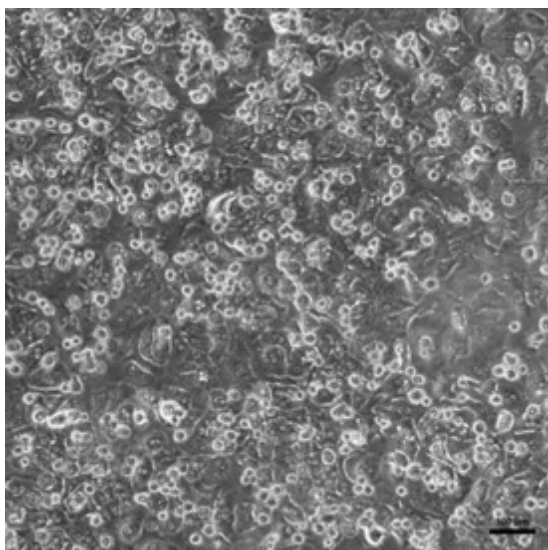
Date: Jan 11, 2024
Amount per vial: 6.96×10^6 cells

Thawing: n=1

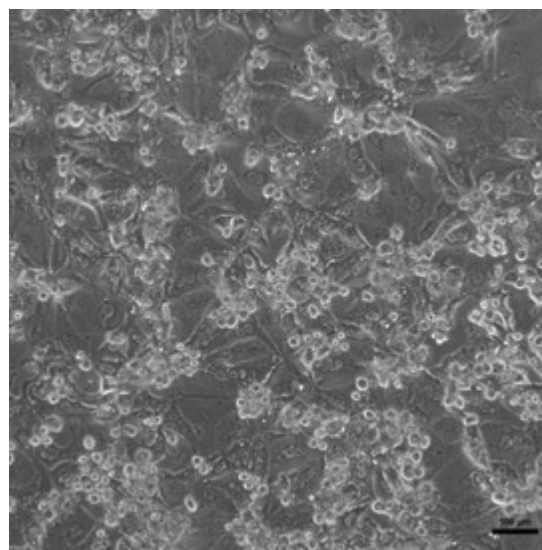
Post-thaw viability: 85.4 %
Post-thaw yield per vial: 2.79×10^6 cells
Recovery: 40 %

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 – 120,000 cells/well // 96well plate – 25,000 cells/well
Culture in Human Hepatocyte Maintenance Medium (HHMM).

CYP P450 activity in 2D culture after thawing:
Ethoxyresorufin-O-deethylation:
Induction with 25 μ M β -Naphthoflavone

pmol/(mg \times min)
96well: 8.46 ± 0.98

X-fold induction
3.0

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	0.5	1	1.5	2	3	4
Viability (%)	85.4	82.5	74.3	77.3	62.8	53.5	53.3

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	Black 6 (C57BL6JR)
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

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.

Issued by: A. Sattler

Verified by: T. Krimmling