

CHCP-I-3D Cynomolgus Hepatocytes Cryopreserved Plateable for Induction assays and for 3D culture

Cell Specification – Certificate of Analysis (CoA)

Lot CH220929-1A

Batch Release: March 27, 2023

Donor data

Species: *Macaca fascicularis*

Gender: male

Age: 3 years 7 months

Serology: negative for Herpes B virus, SRV, SIV, STLV-1, Filovirus (Ebola-like)

Cryopreservation and Thawing

Cryopreservation:

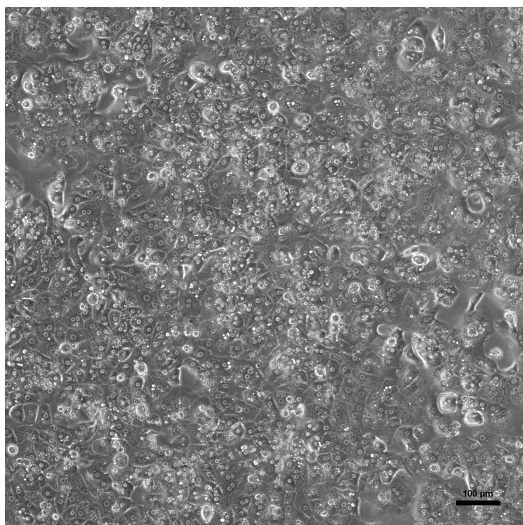
Date: Sept 29, 2022
Amount per vial: 10.0×10^6 cells

Thawing: n=1

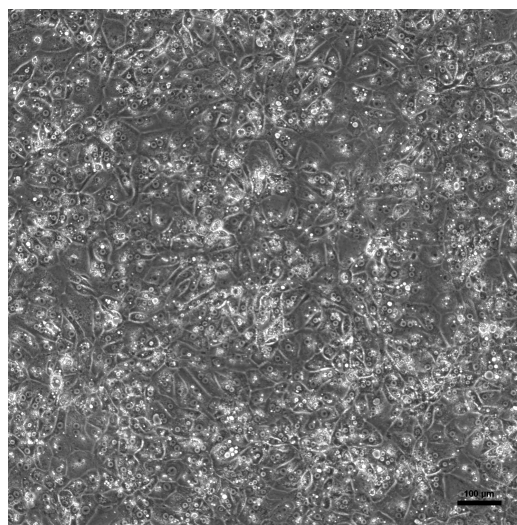
Post-thaw viability: 92.2 %
Post-thaw yield per vial: 7.4×10^6 cells
Recovery: 74 %

2D culture

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

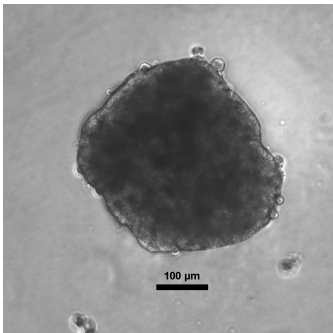
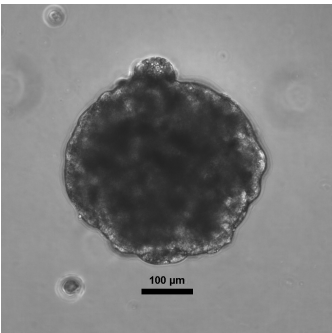


Phase contrast on day 3 after thawing
(24well plate)



Recommended seeding density on collagen-coated plates:
24well plate – 300,000 cells/well // 96well plate – 70,000 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: 57.9 ± 4.7	14.4
Induction with 25 µM β-Naphthoflavone	96well: 99.8 ± 25.0	19.3

3D culture		
Cells seeded in 96well ULA round bottom plates (FaCellitate), 2,500 cells/well		
day 4	day 9	
		
CYP P450 activity in 3D culture after thawing: Ethoxyresorufin-O-deethylation: Induction with 3 µM β-Naphthoflavone	pmol/(mg × min) 5.2 ± 0.8	X-fold induction 2.8

Suspension culture					
Viability test on orbital shaker (Eppendorf Thermomixer C, 1000 rpm at 37 °C with 0.5 x 10 ⁶ cells in 0.5 ml HPM-Cryo):					
Time (h)	0	1	2	3	4
Viability (%)	92.2	74.5	69.6	67.7	60.0
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 <i>in vitro</i> use.					
Issued by: A. Ullrich			Verified by: K. Damrau		