

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

Cell Specification – Certificate of Analysis (CoA)

Lot CH241114 Batch Release: June 02, 2025

Donor data

Gender: male Species: Macaca fascicularis

Age: 5 years 5 months

Serology: negative for Herpes B virus, SRV, SIV, STLV-1

Cryopreservation and Thawing

Cryopreservation:

Date:

Nov 14, 2024

Amount per vial: $13.0 \times 10^6 \text{ cells}$

Thawing: n=2

Post-thaw viability: $96.2 \pm 0.1 \%$

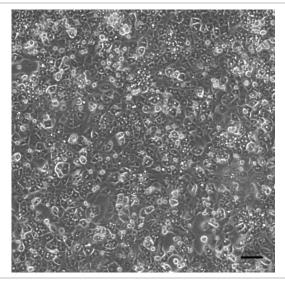
Post-thaw yield per vial: $7.6 \pm 0.2 \times 10^6$ cells

Recovery: $59 \pm 1 \%$

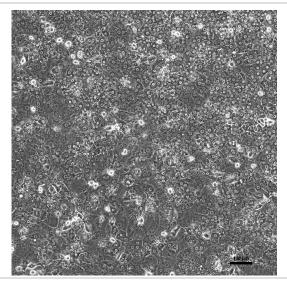
Only one spin required. No washing step.

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 - 60,000 cells/well.

Culture in Human Hepatocyte Maintenance Medium (HHMM).

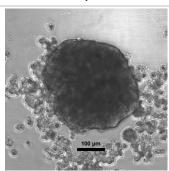
CYP P450 activity in 2D culture after thawing: $pmol/(mq \times min)$ X-fold induction Ethoxyresorufin-O-deethylation: 24well: 72.1 ± 1.3 18.0 Induction with 25 μM β-Naphthoflavone 96well: 111.2 ± 4.1 19.3

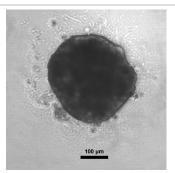


3D culture

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

day 4 day 10





Suspension culture

Viability test on orbital shaker (Eppendorf Thermomixer C, 1000 rpm at 37 $^{\circ}$ C with 0.5 x 10 6 cells in 0.5 ml HPM-Cryo): n=2

Time (h)	0	1	2	3	4	5
Viability (%)	96.2 ± 0.1	91.8 ± 1.0	94.8 ± 3.3	98.1 ± 0.2	99.2 ± 0.8	99.7 ± 0.4

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

Note for thawing process: Only one spin at 100 x g, 10 min., 20 $^{\circ}$ C is required. No washing step needed.

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.

Issued by: A. Ullrich	Verified by: K. Damrau
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