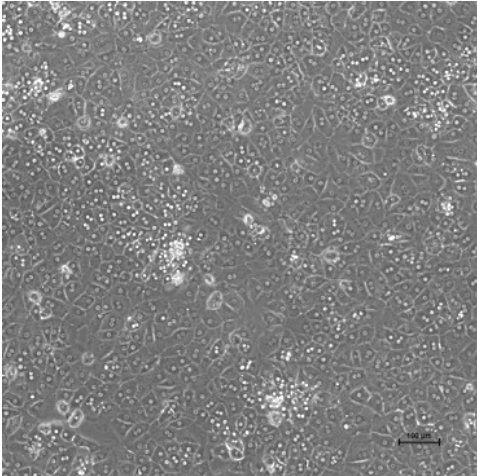
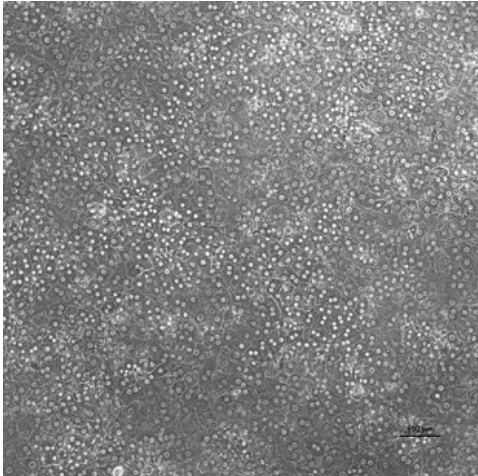


<b>CHCP-I-T Cryopreserved Plateable Cynomolgus Hepatocytes for Induction and Transporter assays</b>		
<b>Cell Specification</b>		
Lot CH140120	Batch Release: May 14, 2019	
Species: <i>Macaca fascicularis</i> Gender: female      Age: 3 years 10 months	Serology: negative for Filovirus/Ebola-like, SRV, SIV, STLV-1	
Cryopreservation: Date:                      January 20, 2014 Amount per vial:      10.2 x 10 <sup>6</sup> cells	Thawing: n=7 Post-thaw viability: 90.3 ± 4.8 % Post-thaw yield per vial: 5.1 ± 0.8 x 10 <sup>6</sup> cells Recovery: 50 %	
Phase contrast on day 1 after thawing	Phase contrast on day 3 after thawing	
		
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 culture after thawing:	pmol/(mg × min)	X-fold induction
Ethoxyresorufin-O-deethylation:	24well: 65.6 ± 0.3	19.9
Induction with 25 µM beta-naphthoflavone	96well: 133.0 ± 18.1	19.7

Uptake Transporters: uptake of 10 $\mu$ M Estrone 3-sulfate (E <sub>3</sub> S) with or without competitive inhibitor Bromosulfophthalein (BSP, 100 $\mu$ M) in cryopreserved hepatocytes after 2 min incubation.		
Activity of uptake transporters in culture after thawing	Intracellular E <sub>3</sub> S (pmol/mg $\times$ min)	Inhibition (%)
Without BSP	371 $\pm$ 102	29.9
With BSP	260 $\pm$ 72	
Efflux Transporter: after 5 min preincubation with 10 $\mu$ M Talinolol (Tal), P-glycoprotein (P-gp) mediated Efflux of Tal with or without the specific P-gp inhibitor PSC833 (10 $\mu$ M) in cryopreserved hepatocytes after 60 min incubation was analyzed.		
Activity of P-gp in culture after thawing	Intracellular Tal (pmol/mg)	
Without PSC833	177 $\pm$ 30	
With PSC833	234 $\pm$ 58	
Note: Yield, viability, recovery and activity assays were performed at PRIMACYT using PRIMACYT's manual for thawing, plating and culture of primary cryopreserved hepatocytes.		
Issued by: A. Ullrich		Checked by: C. Garve