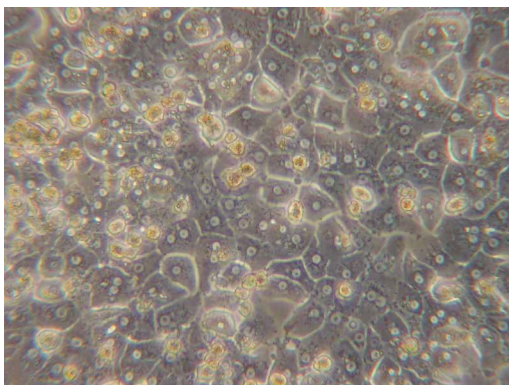
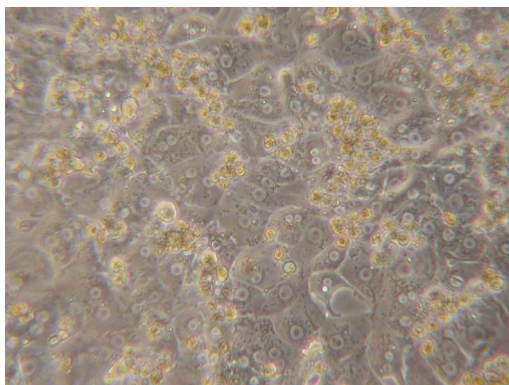


BHCP-I-T Cryopreserved Plateable Beagle Hepatocytes for Induction and Transporter assays
Cell Specification

Lot BH140616-1

Batch Release: November 06, 2014

Species:	Beagle	Gender:	male
		Age:	6 years 5 months
Cryopreservation:		Thawing:	
Date:	June 16, 2014	Post-thaw viability:	75 %
Amount per vial:	10 x 10 ⁶ cells	Post-thaw yield per vial:	4.8 x 10 ⁶ cells
		Recovery:	48 %
Phase contrast on day 1 after thawing		Phase contrast on day 2 after thawing	
			
Recommended seeding density on collagen-coated plates: 283,000 cells per cm ² Culture in Human Hepatocyte Maintenance Medium (HHMM).			
CYP P450 activity in culture after thawing:	pmol/(mg × min)	x-fold induction	
Ethoxyresorufin-O-deethylation:			
Induction with 10 μM beta-naphthoflavone	1.97 ± 0.28	12.3	
Induction with 25 μM beta-naphthoflavone	3.16 ± 0.1	19.8	

Uptake transporters: uptake of 10 µM Estrone 3-sulfate (E₃S) with or without competitive inhibitor Bromosulphophthalein (BSP, 100 µM) in cryopreserved hepatocytes after 2 min incubation.

Activity of uptake transporters in culture after thawing	intracellular E ₃ S (pmol/mg × min)	Inhibition (%)
without BSP	195 ± 54	
with BSP	105 ± 12	46.1

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