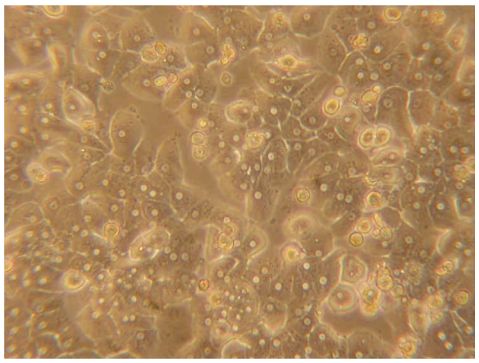
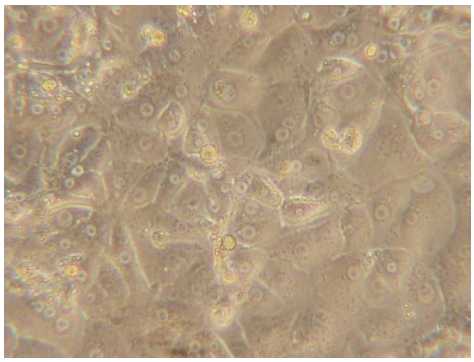
PRIMACYT
Cell Culture Technology

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

Lot BH130510-1-2	Batch Release: November 06, 2014

Species: Beagle	Gender: male Age: 9 months
Cryopreservation: Date: May 10, 2013 Amount per vial: 10.3 x 10^6 cells	Thawing: Post-thaw viability: 71 % Post-thaw yield per vial: 6.2 x 10^6 cells Recovery: 60.2 %
Phase contrast on day 1 after thawing 	Phase contrast on day 3 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	19.9 ± 2.4	25.2
Induction with 25 µM beta-naphthoflavone	33.7 ± 0.7	42.5

Uptake transporters: uptake of 10 µM Estrone 3-sulfate (E_3S) with or without competitive inhibitor Bromosulfophthalein (BSP, 100 µM) in cryopreserved hepatocytes after 2 min incubation.

Activity of uptake transporters in culture after thawing	intracellular E_3S (pmol/mg × min)	Inhibition (%)
without BSP	154 ± 7	
with BSP	93 ± 3	39.6

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