

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

Lot BH140616-2 Batch Release: November 06, 2014

Gender: Species: Beagle male

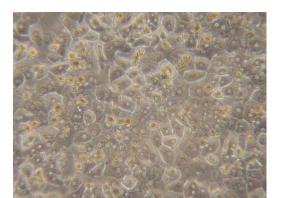
Age: 6 years 5 months Cryopreservation: Thawing:

June 16, 2014 Post-thaw viability: 66 % Date:

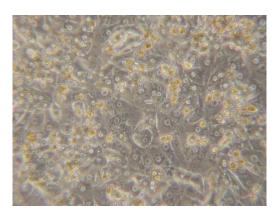
 $10.3 \times 10^6 \text{ cells}$ Post-thaw yield per vial: 4.6 x 10<sup>6</sup> cells Amount per vial:

Recovery: 44.7 %

Phase contrast on day 1 after thawing



Phase contrast on day 2 after thawing



Recommended seeding density on collagen-coated plates: Culture in Human Hepatocyte Maintenance Medium (HHMM).

283,000 cells per cm<sup>2</sup>

CYP P450 activity in culture after thawing:  $pmol/(mg \times min)$ x-fold induction Ethoxyresorufin-O-deethylation:

 $4.09 \pm 0.67$ Induction with 25 µM beta-naphthoflavone 19.6

Uptake transporters: uptake of 10 µM 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 E₃S	Inhibition (%)
	(pmol/mg × min)	
without BSP	163 ± 4	
with BSP	94 ± 26	42.3

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