

Product Characterization Sheet

HH1136

Human Hepatocytes, Catalog Number 82006

Classification

Grade	Highest Quality: 999Elite™
Plateability	Plateable
Viability	≥90%
Confluency	>90%
Culture Duration	>9 Days
P450 Inducibility	Inducible (CYP1A2, CYP2B6, CYP3A4)

Donor Demographics

Gender	Male
Age	16 months
Race	Caucasian
Cause of death	ICH/Stroke
BMI	27.34
Smoking	No
Alcohol	No
Substance abuse	NO
Medical history	Diabetes insipidus
Infectious diseases	HBV-, HCV-, HIV-, CMV+

Post-thaw Viability and Yield

Viability	92 %
Yield	8.5 million

Characterization: Hepatocytes were thawed using 37°C UCRM™ and centrifuged for 10 minutes at 100g. After removing the supernatant, hepatocytes were re-suspended in UPCM™ and counted for viability and yield using the Trypan Blue exclusion method. Cells were plated in a hand-coated collagen 24-well plate at a 0.7 million cells per mL density, 0.5 mL per well, and allowed to attach 4-6 hours prior to a Matrigel® overlay.

P450 Induction

Drug Metabolizing Enzyme	Inducer (µM)	Incubation Time (minutes)	Fold Induction (Gene Expression)
CYP1A2	Omeprazole (50)	30	19.42 ± 0.96
CYP2B6	Phenobarbital (1000)	30	65.42 ± 5.81
CYP2C8	Rifampin (20)	30	
CYP2C9	Rifampin (20)	30	2.33 ± 0.31
CYP2C19	Rifampin (20)	30	0.62 ± 0.07
CYP3A4	Rifampin (20)	30	
	Rifampin (20)	30	10.87 ± 0.43

CYP450 Induction Assessment: 96 well cultures at a cell density of 0.5 million hepatocytes/mL (50,000 hepatocytes/well) were used in the CYP450 induction assessment. The hepatocytes were cultured as collagen-Matrigel® sandwich for 1 day followed by treatment duration of 48-72 hours for mRNA and 72 hours for activity using known enzyme inducers. Induction in CYP450 activity was assessed by quantifying respective metabolite formation by LC-MS/MS. Gene expression was quantified by RT-PCR. Values reflect mean and standard deviation of triplicate treatments (N=3).

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Photomicrographs (100X, Phase Contrast)

Phase Contrast Day 2



Phase Contrast Day 6



Phase Contrast Day 11



Monolayer Comments: It is recommended to seed HH1136 at a seeding density of 0.7 million cells per mL. HH1136 has an excellent attachment efficiency of 100 % and a confluency of 100 % by 24 hours. This lot exhibits excellent morphology and continue to develop a 100 % confluency by day 4. This lot remains intact for over 5 days in culture.

IVAL cell culture media and tissue culture plates used in this evaluation:

- Recovery of thawed hepatocytes - Cat. No. 81015 - UCRM™ Universal Cryopreservation Recovery Media, 50 mL tube
- Initial plating of hepatocytes - Cat. No. 81016 - UPCM™ Universal Primary Cell Plating Media, 50 mL tube
- Sandwich culture with 0.25 mg Matrigel® - Cat. No. 81018/81019 - HIM™ Hepatocyte Induction Media, 50 mL tube/500 mL bottle
- Suspension and incubation of hepatocytes - Cat. No. 81039/81040 - HQM™ Hepatocyte Incubation Media, 50 mL tube/500 mL bottle
- Collagen coated plates - Cat. No. 71006, 71008 - CellAffix™ 24-well and 96-well Collagen Hand Coated tissue culture plate, 5 plates per pack

To inquire about our products and services or for technical questions please contact:

- In Vitro ADMET Laboratories by phone at +1 (866) 458-1094 or +1 (410) 869-9037 or email at info@invitroadmet.com