

Product Characterization Sheet

HH1110

Human Hepatocytes, Catalog Number 82006



Classification

Plateability	Plateable
Number of days plateable	1 day
Confluency	70 %
P450 Inducibility	No
Transporter activity	No
Number of donors	2

Donor Demographics

Gender	Male
Age	47
Race	Hispanic
Cause of death	CVA 2 nd to ICH
BMI	25.2
Smoking	Yes
Alcohol	Yes
Substance abuse	No
Medical history	Diabetes, HTN,
Infectious diseases	HBV-, HCV-, HIV-, CMV+, EBV (IgG)+

Post-thaw Viability and Yield

Viability	83 %
Yield	6.1 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.

Drug Metabolism Activity

Drug Metabolizing Enzyme	Substrate (μM)	Incubation Time (minutes)	Metabolite Quantified	Activity (pmol/minute/million cells)
CYP1A2	Phenacetin (100)	15	Acetaminophen	20 ± 2.5
CYP2A6	Coumarin (50)	30	7-Hydroxycoumarin	234.5 ± 20.4
CYP2B6	Bupropion (500)	15	Hydroxybupropion	35.1 ± 3.4
CYP2C8	Paclitaxel (20)	15	6α-Hydroxypaclitaxel	51.6 ± 5
CYP2C9	Diclofenac (25)	15	4-Hydroxydiclofenac	137 ± 8
CYP2C19	S-Mephenytoin (250)	30	4-Hydroxymephenytoin	3 ± 0.9
CYP2D6	Dextromethorphan (15)	15	Dextrorphan	10.8 ± 1.2
CYP2E1	Chlorzoxazone (250)	15	6-Hydroxychlorzoxazone	30 ± 5.4
CYP3A4	Midazolam (20)	10	1-Hydroxymidazolam	14 ± 0.6
	Testosterone (200)	15	6β-Hydroxytestosterone	116 ± 40.8
ECOD	7-Ethoxycoumarin (100)	30	7-Hydroxycoumarin	121.7 ± 1.1
UGT	7-Hydroxycoumarin (100)	30	7-Hydroxycoumarin glucuronide	664 ± 43
Sulfotransferase	7-Hydroxycoumarin (100)	30	7-Hydroxycoumarin sulfate	34.6 ± 5.3

CYP450 Activity Assessment: The hepatocytes were incubated at a cell density of 0.5 million cells/mL in a 48-well plate (125,000 hepatocytes/well) for the designated time durations with isoform-selective substrates. The metabolites were identified and analyzed using LC-MS/MS.

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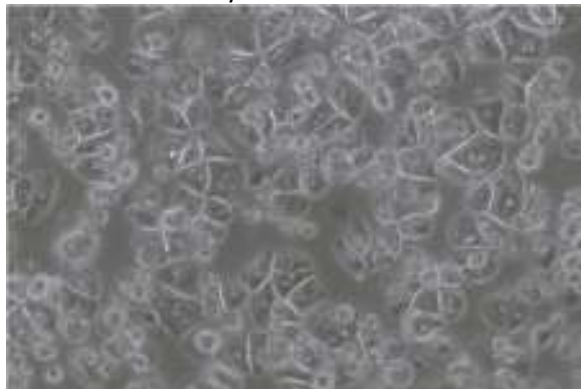
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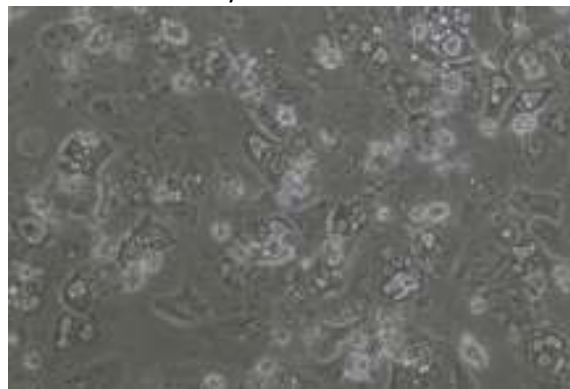


Photomicrographs (100X, Phase Contrast)

Phase Contrast Day 2



Phase Contrast Day 5



Monolayer Comments: HH1110 has a good attachment efficiency and a confluency of 70 % by 24 hours. HH1110 attachment is improved by plating hepatocytes at a density of 1.0 million cells per mL. This lot exhibits deterioration of the monolayer by day 3 and is recommended for short term 2 day studies.

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