

# Product Characterization Sheet

## HH1043

Human Hepatocytes, Catalog Number 82006



### Classification

Plateability	Plateable
Number of days plateable	5 days
Confluency	60 %
P450 Inducibility	No
Transporter activity	No
Number of donors	1

### Donor Demographics

Gender	Male
Age	37 years
Race	Hispanic
Cause of death	CVA 2 <sup>nd</sup> to ICH
BMI	41.7
Smoking	Yes
Alcohol	Yes
Substance abuse	Yes
Medical history	NA
Infectious diseases	HBV-, HCV-, HIV-, CMV+

### Post-thaw Viability and Yield

Viability	89 %
Yield	6.7 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	50.8
CYP2A6	Coumarin (50)	30	7-Hydroxycoumarin	39.8
CYP2B6	Bupropion (500)	15	Hydroxybupropion	8.1
CYP2C8	Paclitaxel (20)	15	6α-Hydroxypaclitaxel	8.5
CYP2C9	Diclofenac (25)	15	4-Hydroxydiclofenac	23.3
CYP2C19	S-Mephenytoin (250)	30	4-Hydroxymephenytoin	4.7
CYP2D6	Dextromethorphan (15)	15	Dextrorphan	17.7
CYP2E1	Chlorzoxazone (250)	15	6-Hydroxychlorzoxazone	18.9
CYP3A4	Midazolam (20)	10	1-Hydroxymidazolam	10.5
	Testosterone (200)	15	6β-Hydroxytestosterone	180.0
ECOD	7-Ethoxycoumarin (100)	30	7-Hydroxycoumarin	60.2
UGT	7-Hydroxycoumarin (100)	30	7-Hydroxycoumarin glucuronide	301.7
Sulfotransferase	7-Hydroxycoumarin (100)	30	7-Hydroxycoumarin sulfate	48.4

**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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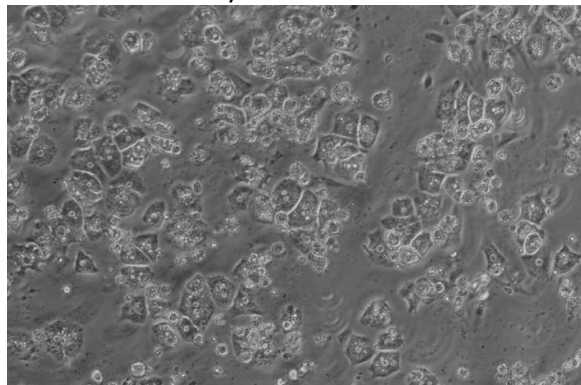
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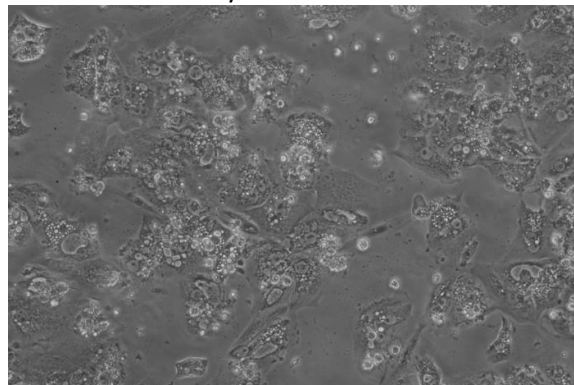


### Photomicrographs (100X, Phase Contrast)

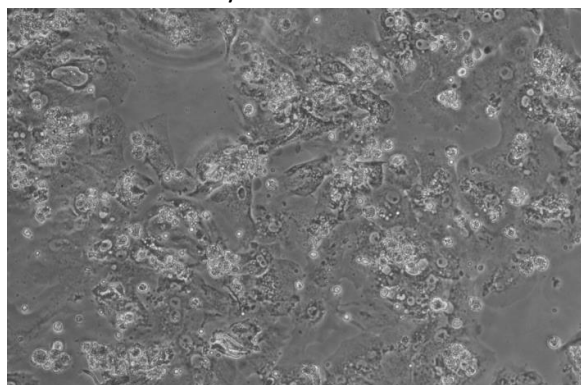
Phase Contrast Day 2



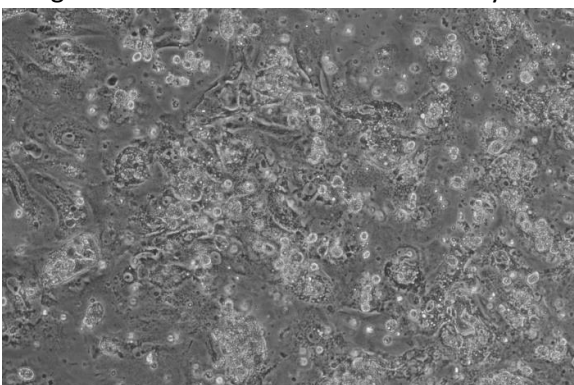
Phase Contrast Day 4



Phase Contrast Day 5



Long-term Human Plasma Incubation Day 7



**Monolayer Comments:** HH1043 has a fair attachment efficiency and a confluency of 60 % by 24 hours. Stretching of hepatocytes is observed and the lot remains intact for 5 days in culture.

**Human Plasma Incubation:** The hepatocytes were cultured at a cell density of 0.35 million hepatocytes/ 0.5 mL in a 24-well plate as a collagen-Matrigel® sandwich. On day 2, the hepatocytes were treated with media containing human plasma and received subsequent media changes every other day to assess long term effects of plasma on morphology.

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](mailto:info@invitroadmet.com)