Product Characterization Sheet HE3047





Classification

Plateability	Suspension			
Number of days plateable	NA			
Confluency	NA			
P450 Inducibility	NA			
Number of donors	1			
Enterocyte size	16.9 ± 3.4			

Post-thaw Viability and Yield

Viability	81 %	
Yield	3.1 million	

Donor Demographics

0 1		
Gender	Male	
Age	55 years	
Race	Caucasian	
Cause of death	CVA 2 nd to ICH	
вмі	24.4	
Smoking	No	
Alcohol	No	
Substance abuse	No	
Medical history	HTN, depression, GERD, bipolar, vascular dimentia	
Infectious diseases	HBV-, HCV-, HIV-, CMV+, EBV (IgG)+	

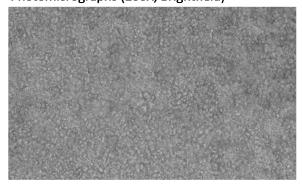
<u>Characterization:</u> Enterocytes were thawed using 37° C CERMTM and centrifuged for 10 minutes at 100g. After removing the supernatant, enterocytes were re-suspended in 4° C HQMTM and counted for viability and yield using the Trypan Blue exclusion method.

Drug Metabolism Activity

Drug Metabolizing Enzyme	Substrate (μM)	Incubation Time (minutes)	Metabolite Quantified	Activity (pmol/minute/million cells)
СҮРЗА4	Midazolam (20)	60	1-Hydroxymidazolam	0.55 ± 0.01
	Testosterone (200)	60	6β-Hydroxytestosterone	8 ± 0.58
CYP2J2	Astemizole (50)	60	O-Desmethyl astemizole	0.26 ± 0.01
CES2	Irinotecan (50)	60	SN38	0.51 ± 0.04
CYP2C9	Diclofenac (25)	60	4-Hydroxydiclofenac	1.24 ± 0.1
CYP2C19	S-Mephenytoin (250)	60	4'-Hydroxymephenytoin	0.08 ± 0.06
Sulfotransferase	7-Hydroxycoumarin (100)	60	7-Hydroxycoumarin sulfate	5.1 ± 0.03
UGT	7-Hydroxycoumarin (100)	60	7-Hydroxycoumarin glucuronide	2.9 ± 0.1

<u>CYP450 Activity Assessment:</u> The enterocytes were incubated at a cell density of 1.5 million enterocytes/mL in a 96-well plate (150,000 enterocytes/well) for the designated time duration of 120 minutes with isoform-selective substrates. The metabolites were identified and analyzed using LC-MS/MS.

Photomicrographs (100X, Brightfield)



Enterocyte Morphology: Enterocyte cell size is approximately 12 – 17 microns in diameter. Lymphocytes are approximately 4 microns and may also be observed in the enterocyte cell population.

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

- Recovery of thawed enterocytes Cat. No. 81056 CERM™ Cryopreserved Enterocyte Recovery Media, 50 mL tube
- Suspension and incubation of enterocytes Cat. No. 81039 HQM™ Hepatocyte and Enterocyte Incubation Media, 50 mL tube

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