

Updated on: 28th October 2024

CERTIFICATE OF ANALYSIS

Lot#: CHM2420-L-HE-Z

PRODUCT DESCRIPTION

Reference: HuHECP/4+ Product: Cryopreserved Human Hepatocytes Category: Plateable Spheroid qualified: Yes Specific culture requirements: No Isolation date: 17th October 2024 Initial Isolation Viability: 92.00% Storage conditions: -196°C using LN₂ Sterility test: Negative for mycoplasma, bacteria, yeast, and fungi

DONOR DEMOGRAPHICS

Species	Gender	Race	Age	BMI	Smoker	Alcohol Use	Drug Use		
Human	Male	Caucasian	an 54 23.5 N/A N/A				N/A		
Pathology			Serological Data ¹						
Hepatic metastases				Tested negative less than 3 months before surgery					

Patient informed consent was obtained. ¹The donor was serologically tested negative for following infectious diseases: HIV, Hepatitis B and C, and SARS-CoV-2. Donor medical history was also examined prior to accepting this donor. *For donor's medication information, please contact us.*

CHARACTERIZATION FOR PLATEABLE CELLS

Post Thaw Lot information	Result	SD	n
Number of viable cells (cells/vial):	5.36x10 ⁶	± 0.23x10 ⁶	2
Post-thaw viability (%):	88.42	± 1.07	2
Days in culture after thaw (24w):	N/A	± 0.00	1
Days in culture after thaw (96w):	N/A	± 0.00	1
MONOLAYER ASSESSMENT ² Plateable:	YES Conflu	uence 24h: 95%	
Seeding density in 24 well recommended:	2.10>	د10 ⁵ cells/cm ²	
Seeding density in 96 well recommended:	2.81>	(10 ⁵ cells/cm ²	
Cell morphology 24h		Cell morphology S	96h
	50 µm		С. С

Human hepatocytes were thawed and seeded according to BeCytes Biotechnologies culture protocol. The yield and viability were determined by a trypan blue exclusion assay after the thawing process. ²Resuspended human hepatocytes from post-thaw assessment were plated in collagen-coated 24-well plates in hepatocyte plating medium. Cells were refreshed with hepatocytes maintenance

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medium during the first change of medium on the day of thawing. Maintenance medium was replaced in the culture every day. If images from the 96-well plates are needed, please contact us.

3D SPHEROID FORMATION

Spheroid morphology



Primary human hepatocytes self-assembled into a spheroid containing 1000 cells after 4 days in culture. These hepatic spheroids were cultured for 7-15 days in ultra-low attachment (ULA) plates with our 3D Culture Maintenance Media for hepatocytes (MHM3D). For more information/protocols about 3D hepatocyte spheroids, contact us.

If you need help for an experiment, just contact us, our experts will be pleased to assist you

CERTIFICATION:

The viability and performance of the primary human hepatocytes provided depend primarily on the use of appropriate media and reagents, as well as the use of sterile plastics. Likewise, proper handling protocols must be followed. Please note that if these parameters are not carefully considered, the cellular response obtained in the assays may be lower than expected.

Name	Tittle	Signature	Cytes Biotechnologies, S.L.	Date
Pilar Sainz de la Maza	Quality Manager	Play four level	CYTES BIOTECHICA COGIES SL	28/10/2024

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CELL COUNTING

Lot #:_____

Date: ___/___/

MORPHOLOGY

Clear cytoplasmClear membranes

Rounded shapeMembrane blebbing

Cell swelling
 Lipid droplets

Hardly any debrisPrevalent debris

TRYPAN BLUE COUNTING RESULTS

$\frac{1}{1} \frac{1}{1} \frac{1}$	NEUBAUER CHAMBER COUNTING								
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COMMENTS	COMMENTS								

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