

CERTIFICATE OF ANALYSIS

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Cell Line Details

Product code: HEP-003-OTC-a

Product name: iPSC-derived Human Hepatocyte-Like Cells: CRISPR engineered Hemizygous Ornithine Transcarbamylase; OTC D175V

Lot number: XXXXXX

Storage conditions: Store at less than -130°C

QC completion date: XXXXXX

Cell Quality Controls

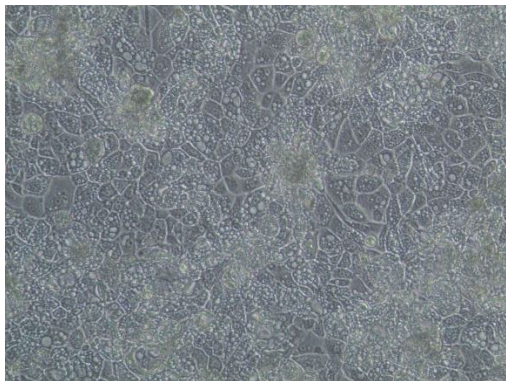
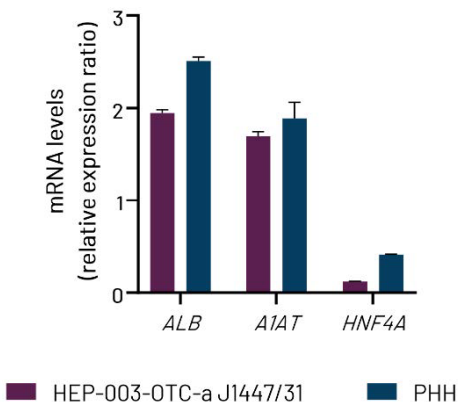
Test	Method	Specification	Result
Virus test for original iPSC clone (HIV1, HIV2, Hepatitis A, HBV, HCV, HTLV-1, HTLV-2)	PCR	Not detected	Pass
Mycoplasma	qPCR	Not detected	Pass
Post thaw viability	Automated cell counter	≥ 70% viable	Pass
Viable cells per vial	Automated cell counter	≥ 5.0 x 10 ⁶	Pass
Cell morphology	Visual check	N/A	Pass (Fig.1)
Key hepatocyte maturity markers (<i>ALB</i> , <i>A1AT</i> , <i>HNF4a</i>)	qPCR	Present	Pass (Fig.2)
Disease markers (OTC)	Western blot	Decreased OTC protein expression	Pass (Fig.3)
Disease confirmation	Sanger sequencing	Mutation present	Pass (Fig.4)

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Appendix

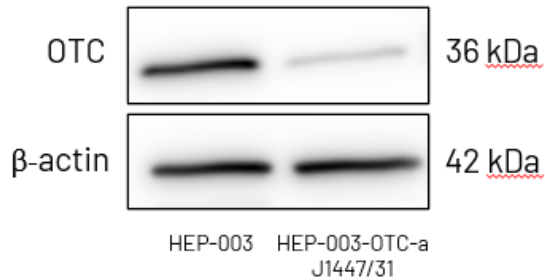
<p>1. Cell morphology</p> <p>Figure 1. Morphology of cryopreserved hepatocyte-like cells, 14 days post-thaw. Brightfield picture, objective: 10x.</p>													
<p>2. Detection of hepatocyte maturity markers via qPCR</p> <p>Figure 2. Gene expression of key hepatocyte maturity markers: ALB (Albumin), A1AT (Alpha-1 Antitrypsin), and HNF4A (Hepatocyte Nuclear Factor-4) in cryopreserved hepatocyte-like cells and primary human hepatocytes (PHH), 14 days post-thaw. mRNA data are normalized to endogenous PPIA expression, and are presented as mean±SD of n=3 technical replicates.</p>	 <table><tr><th>Marker</th><th>HEP-003-OTC-a J1447/31</th><th>PHH</th></tr><tr><td>ALB</td><td>~2.0</td><td>~2.5</td></tr><tr><td>A1AT</td><td>~1.8</td><td>~2.0</td></tr><tr><td>HNF4A</td><td>~0.2</td><td>~0.5</td></tr></table>	Marker	HEP-003-OTC-a J1447/31	PHH	ALB	~2.0	~2.5	A1AT	~1.8	~2.0	HNF4A	~0.2	~0.5
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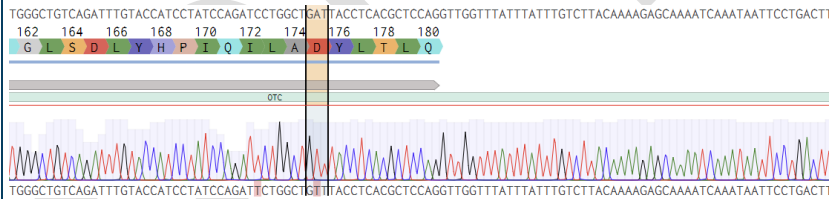
3. Disease confirmation

Figure 3. Protein expression levels of OTC and β -actin in wild-type (HEP-003) and OTC deficiency (HEP-003-OTC-a) cells analyzed by western blot.



4. Disease confirmation via Sanger sequencing

Figure 4. Sanger sequencing showing hemizygous mutation D175V (GAT>GTT) within OTC gene (exon 5) in HEP-003-OTC-a cells. Wild-type gene sequence is shown on top.



Checked by,

signature

QC Scientist

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