HepatoXcell™ Pro: Normal Human Hepatocytes

PCS-450-011™

Description

HepatoXcell™

HepatoXcell™ Pro are Primary Human Hepatocytes, 7 day Plateable, derived from normal, healthy, human liver tissues.

Organism: *Homo sapiens*, human
Tissue: Liver
Morphology: polygonal epithelial cells with one or multiple nuclei while in adherent cell culture
Growth properties: Adherent
Cells per vial: ≥ 4.0 x 10^6

Storage Conditions

Product format: Frozen
Storage conditions: Vapor phase of liquid nitrogen

Intended Use

This product is intended for laboratory research use only. It is not intended for any animal or human therapeutic use, any human or animal consumption, or any diagnostic use.

BSL 2

ATCC determines the biosafety level of a material based on our risk assessment as guided by the current edition of *Biosafety in Microbiological and Biomedical Laboratories (BMBL)*, U.S. Department of Health and Human Services. It is your responsibility to understand the hazards associated with the material per your organization's policies and procedures as well as any other applicable regulations as enforced by your local or national agencies.

ATCC highly recommends that appropriate personal protective equipment is always used when handling vials. For cultures that require storage in liquid nitrogen, it is important to note that some vials may leak when submersed in liquid nitrogen and will slowly fill with liquid nitrogen. Upon thawing, the conversion of the liquid nitrogen back to its gas phase may result in the vial exploding or blowing off its cap with dangerous force creating flying debris. Unless necessary, ATCC recommends that these cultures be stored in the vapor phase of liquid nitrogen rather than submersed in liquid nitrogen.

Certificate of Analysis

For batch-specific test results, refer to the applicable certificate of analysis that can be found at www.atcc.org.

Growth Conditions

Temperature: 37°C
Atmosphere: 95% Air, 5% CO_2

Handling Procedures

Complete medium: The complete media used are HepatoXcell™ Primary Hepatocyte Plating Medium 1x (ATCC PCS-450-038) and HepatoXcell™ Primary Hepatocyte Maintenance Medium 1x (ATCC PCS-450-034).

Handling Procedure:

Unpacking and storage instructions

- Check all containers for leakage or breakage.
- Remove the frozen cells from the liquid nitrogen dry shipper and immediately place the cells at a nitrogen vapor dewar with a temperature below -130°C, until ready for use.

Required media and supplement

• One bottle of each of the following: Hepatocyte Thaw Media (ATCC PCS-450-032), Hepatocyte Plating Media (ATCC PCS-450-038), Hepatocyte Maintenance Media (ATCC PCS-450-034). Cell Basement Membrane (ATCC ACS-3035). Optional media supplements: Penicillin-Streptomycin-Amphotericin B Solution (ATCC PCS-999-002)

Handling procedure

1. Refer to the batch specific information for the total number of viable cells recovered post-thaw for any lot of PCS-450-011.
2. Add 19 mL of pre-warmed Hepatocyte Thaw Media to a 50 mL centrifuge tube.
3. Remove one vial of PCS-450-011 from storage and thaw the cells by gentle agitation in a 37°C water bath. To reduce the possibility of contamination, keep the O-ring and cap out of the water. Thawing should be rapid (approximately 1 to 2 minutes).
4. Remove the vial from the water bath as soon as the contents are thawed leaving a small ice pellet and decontaminate by dipping in or spraying with 70% ethanol. All operations from this point onward should be carried out under strict aseptic conditions.
5. Gently pour the contents of the vial into the 50 mL centrifuge tube.
6. Using a wide bore pipette tip wash the vial with 1 mL of the Hepatocyte Thaw Media suspension to retrieve any cells left in the vial.

Note: When pipetting Hepatocytes NEVER pipette up and down to mix, instead gently rock or shake the tube.

7. Centrifuge at 100 x g for 10 minutes.
8. Carefully remove the supernatant and resuspend with 1 mL of prewarmed Hepatocyte Plating Media.
9. Gently rock or shake the tube to resuspend the cells, then add an additional 1 mL

of Hepatocyte Plating Media.
10. Mix gently by rocking or shaking to ensure a homogenous suspension.
11. Perform a cell count using the Trypan Blue Exclusion Method.
12. Using the total number of viable cells, dilute the cells to 800,000 cells/mL.
13. Seed one well in a Collagen I coated 24-well plate with 500 µL of the cell suspension.
14. Gently shake plate in a “T” motion to evenly distribute the cells.
15. Under the microscope, let the cells settle to ensure seeding density is not too low or high.
16. If the seeding density is too low add 50 µL of cell suspension to the well, if too high shake the plate to resuspend cells and remove 50 µL of cell suspension.
17. After seeding density is optimized, seed remaining wells with Human Hepatocyte cell suspension.
18. Gently shake in a “T” motion and place into a 36°C ± 2°C, 5% CO2 ± 2% CO2 incubator.
19. At 1 hour post seeding gently shake the plate in a “T” motion to evenly distribute the cells and remove any dead cells attached to the plate wells.
20. At 2 hours post-seeding gently shake the plate in a “T” motion and perform a media change using pre-warmed plating media to remove excess dead cells.
21. Four to six hours post-seeding, perform a media change with cold Hepatocyte Maintenance Media supplemented with 0.3 mg/mL Cell Basement Matrix.
22. Put plate back into incubator and change media daily.

Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: HepatoXcell™ Pro: Normal Human Hepatocytes (ATCC PCS-450-011)

References

References and other information relating to this material are available at www.atcc.org.

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