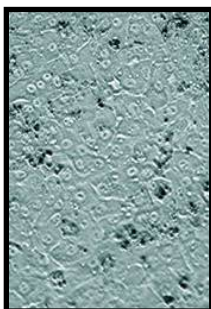


HEPATIC CELL SYSTEM INNOPROFILE™ HUMAN HEPATOCYTES



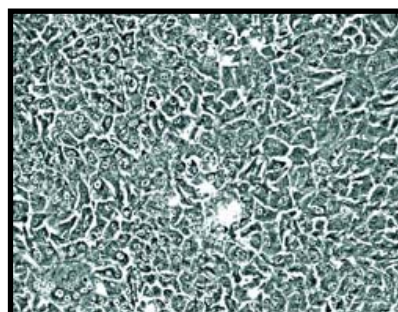
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|-------------------------|--|
| Product Type: | Cryo-preserved Hepatocytes |
| Catalog Number: | P10651 |
| Source: | Human Liver |
| Number of Cells: | 1 x 10 ⁶ Cells / vial (1ml) |
| Storage: | Liquid Nitrogen |

Human Hepatocytes (HH) provided by Innoprot are isolated from human healthy liver. HH are cryopreserved immediately after purification and delivered frozen. HH are guaranteed to further culture in the condition provided in this technical sheet.

The liver fulfills many vital processes in mammals. It is the central organ of energy metabolism, biotransformation of xenobiotics, and synthesis of plasma proteins under physiological and pathophysiological conditions. Primary culture of human hepatocyte appears to be a suitable experimental model for the study of liver specific function, have been and still is an important tool. Propagation of human hepatocytes for cell transplantation, gene therapy, and culture of hepatocyte in bioartificial liver support systems is now under investigation. In appropriate culture condition, cultured human hepatocytes proliferate and maintain differentiated hepatocyte function such as the synthesis of serum proteins.

Recommended Medium

- Hepatocyte Medium
(Reference: P60109)



Product Characterization

Immunofluorescent method

- Cytokeratin-18
- Vimentin
- Albumin

The cells test negative for HIV-1, HBV, HCV, mycoplasma, bacteria, yeast and fungi

Product Use

THESE PRODUCTS ARE FOR RESEARCH USE ONLY. Not approved for human or veterinary use, for application to humans or animals, or for use in vitro diagnostic or clinical procedures

INSTRUCTIONS FOR CULTURING CELLS

IMPORTANT: Experiments should be well organized before thawing cells, as hepatocytes do not proliferate in culture. It is recommended that HH are used for experiments as quickly as possible after thawing the cells

Set up culture after receiving the order:

1. Prepare a fibronectin coated flask (2 $\mu\text{g}/\text{cm}^2$, T-75 flask is recommended). Add 10 ml of sterile Dulbecco's phosphate buffered saline (DPBS) to a T-75 flask and then add 150 μl of fibronectin solution (1 mg/ml, Ref:P8248). Leave the flask in incubator overnight.
2. Prepare complete medium. Decontaminate the external surfaces of medium bottle and medium supplement tubes with 70% ethanol and transfer them to a sterile field. Aseptically transfer supplement to the basal medium with a pipette. Rinse the supplement tube with medium to recover the entire volume.
3. Rinse the fibronectin-coated vessel twice with sterile water and then add 15 ml of complete medium. Leave the vessel in the sterile field and proceed to thaw the cryopreserved cells.
4. Place the frozen vial in a 37°C water bath. Hold and rotate the vial gently until the contents completely thaw. Promptly remove the vial from the water bath, wipe it down with 70% ethanol, and transfer it to the sterile field.
5. Carefully remove the cap without touching the interior threads. Gently resuspend and dispense the contents of the vial into the equilibrated, fibronectin-coated culture vessel. A seeding density higher than 20,000 cells/cm² is recommended.

Note: Dilution and centrifugation of cells after thawing are not recommended since these actions are more harmful to the cells than the effect of residual DMSO in the culture. It is also important that cells are plated in fibronectin-coated culture vessels to promote cell attachment.

6. Replace the cap or lid of the culture vessel and gently rock the vessel to distribute the cells evenly. Loosen cap, if necessary, to allow gas exchange.
7. Return the culture vessel to the incubator.
8. For best results, do not disturb the culture for at least 16 hours after the culture has been initiated. Refresh culture medium the next day to remove residual DMSO and unattached cells.
9. Use cells promptly for experiments.

Note: HH cannot be subcultured or passaged since this cell type will terminally differentiate in long term culture.

Caution: Handling human derived products is potentially biohazardous. Although each cell strain testes negative for HIV, HBV and HCV DNA, diagnostic tests are not necessarily 100% accurate, therefore, proper precautions must be taken to avoid inadvertent exposure. Always wear gloves and safety glasses when working these materials. Never mouth pipette. We recommend following the universal procedures for handling products of human origin as the minimum precaution against contamination [1].