Human Hepatocytes (HH)

Catalog Number: 5200

Cell Specification

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 [1]. 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 [2]. In appropriate culture condition, cultured human hepatocytes proliferate and maintain differentiated hepatocyte function such as the synthesis of serum proteins [3].

HH from ScienCell Research Laboratories are isolated from human liver. HH are cryopreserved immediately after purification and delivered frozen. Each vial contains >1 x 10^6 cells in 1 ml volume. HH are characterized by immunofluorescent method with antibodies to albumin, cytokeratin-18 and vimentin. HH are negative for HIV-1, HBV, HCV, mycoplasma, bacteria, yeast and fungi. *HH is not recommend for expanding or long term cultures since the cells do not proliferate in culture*.

Recommended Medium

It is recommended to use Hepatocyte Medium (HM, Cat. No. 5201) for the culturing of HH *in vitro*.

Product Use

HH are for research use only. It is not approved for human or animal use, or for application in *in vitro* diagnostic procedures.

Storage

Directly and immediately transfer cells from dry ice to liquid nitrogen upon receiving and keep the cells in liquid nitrogen until cell culture is needed for experiments.

Shipping

Dry ice.

Reference

[1] Runge, D., Michalopoulos, G. K., Strom, S. C. and Runge, D. M. (2000) Recent advances in human hepatocyte culture systems. *Biochem. Biophysi. Res. Comm*. 274:1-3.

[2] Chen, H. L., Wu, H. L., Fon, C. C., Chen, P. J., Lai, M. Y. and Chen, D. S. (1998) Long-term culture of hepatocytes from human adults. *J. Biomed. Sci*. 5:435-440.

[3] Okamoto, M., Ishida, Y., Keogh, A. and Strain, A. (1998) Evaluation of the function of primary human hepatocytes co-cultured with the human hepatic stellate cell (HSC) line LI90. *Int. J. Artif. Organs* 21:353-359.

Instruction for culturing cells

Caution: Cryopreserved cells are very delicate. Thaw the vial in a 37°C waterbath and return them to culture as quickly as possible with minimal handling!

Set up culture after receiving the ordering:

1. Prepare a poly-L-lysine coated flask (2 μg/cm^2, T-75 flask is recommended). Add 10 ml of sterile water to a T-75 flask and then add 15 μl of poly-L-lysine stock solution (10 mg/ml, ScienCell cat. no. 0413). Leave the flask in incubator overnight (minimum one hour at 37°C incubator).
2. Prepare complete medium: decontaminate the external surfaces of medium and medium supplements with 70% ethanol and transfer them to sterile field. Aseptically open each supplement tube and add them to the basal medium with a pipette. Rinse each tube with medium to recover the entire volume.
3. Rinse the poly-L-lysine coated flask with sterile water twice and add 20 ml of complete medium to the flask. Leave the flask in the hood and go to thaw the cells.
4. Place the vial in a 37°C waterbath, hold and rotate the vial gently until the contents are completely thawed. Remove the vial from the waterbath immediately, wipe it dry, rinse the vial with 70% ethanol and transfer it to a sterile field. Remove the cap, being careful not to touch the interior threads with fingers. Using a 1 ml eppendorf pipette gently re-suspend the contents of the vial.
5. Dispense the contents of the vial into the equilibrated, poly-L-lysine coated culture vessels. A seeding density of 5,000 cells/cm^2 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 DMSO residue in the culture. It is also important that cells are plated in poly-L-lysine coated flask that promotes cell attachment and growth.
6. Replace the cap or cover, and gently rock the vessel to distribute the cells evenly. Loosen cap if necessary to permit gas exchange.
7. Return the culture vessels to the incubator.
8. For best result, do not disturb the culture for at least 16 hours after the culture has been initiated. Change the growth medium the next day to remove the residual DMSO and unattached cells, then every other day thereafter.

Maintenance of Culture:

1. Change the medium to fresh supplemented medium the next morning after establishing a culture from cryopreserved cells.

2. Change the medium every two to three days thereafter.

HH are not recommended to be subcultured since this cell type will terminally differentiate in long term culture.

Caution: Handling human derived products is potentially biohazardous. Although each cell strain tests 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].

[1]. Grizzle, W. E., and Polt, S. S. (1988) Guidelines to avoid personal contamination by infective agents in research laboratories that use human tissues. J Tissue Culture Methods. 11(4).