

Product Information

Human Hepatic Stellate Cells - NASH (HHSC -N)

Catalog Number	10HU-210N	Cell Number	1.0 million cells/vial
Species	<i>Human</i>	Storage Temperature	Liquid nitrogen

Description

Hepatic stellate cells (HSCs) are liver-specific mesenchymal cells, and account for 5–8% of the cells in the liver. HSCs play vital roles in the homeostasis of liver extracellular matrix, repair, regeneration and fibrosis, and control retinol metabolism, storage and release. The stellate cell is the major cell type involved in liver fibrosis in response to liver injury.

In healthy liver, HSCs are in a quiescent state, and contains numerous vitamin A lipid droplets, constituting the largest reservoir of vitamin A in the body. When the liver is damaged, HSCs can change into an activated state, which is characterized by proliferation, contractility and chemotaxis. The amount of vitamin A decreases progressively in injured liver. The activated HSCs also secrete collagen scar tissue, which can lead to cirrhosis. In chronic liver disease, prolonged and repeated activation of stellate cells causes liver fibrosis ^[1,2].

In non-alcoholic steatohepatitis (NASH), hepatic stellate cells (HSC) differentiate into myofibroblast-like cells that cause fibrosis, which predisposes patients to cirrhosis and hepatocellular carcinoma. Thus, modeling interactions between activated HSCs and hepatocytes in vitro can aid in the development of anti-NASH/fibrosis therapeutics and lead to a better understanding of disease progression ^[3].

iXCells Biotechnologies provides high quality Human Hepatic Stellate Cells - Nash (HHSC -N), which are isolated from liver tissue of NASH patient, and cryopreserved with >1 million cells in each vial. Each vial is sufficient to seed on four 10 cm dishes, and cells will be ready in 5–6 days to be passaged at 1:3 ratio. HHSC express Desmin and GFAP, and are negative for HIV-1, HBV, HCV, mycoplasma, bacteria, yeast, and fung. HHSC can further expand for 2-3 population doublings in Stellate Cell Growth Medium (Cat # MD-0014) under the condition suggested by iXCells Biotechnologies.

Product Details

Tissue	Adult human liver from NASH patient
Package Size	1.0 million cells/vial
Shipped	Cryopreserved
Storage	Liquid nitrogen
Growth Properties	Adherent
Media	Stellate Cell Growth Medium (Cat# MD-0014)

Protocols

Thawing of Frozen Cells

1. Upon receipt of the frozen Human Hepatic Stellate Cells (HHSC), it is recommended to thaw the cells and initiate the culture immediately in order to retain the highest cell viability.
2. To thaw the cells, put the vial in 37°C water bath with gentle agitation for ~1 minute until just the tiniest ice crystal is left in the cryovial. Keep the cap out of water to minimize the risk of contamination.
3. It is recommended to seed one vial of HHSCs into four 100mm cell culture dishes in Stellate Cell Growth Medium (Cat # MD-0014).

Note: Do not use cell culture flasks, because HHSCs may become activated.

4. Change the medium every other day until the cells reach 80-90% confluence.

Safety Precaution: it is highly recommended that protective gloves and clothing should be used when handling frozen vials.

Sub-culture Procedure

1. Human Hepatic Stellate Cells (HHSC) can be cultured in Stellate Cell Growth Medium (Cat # MD-0014).
2. When cells reach ~80-90% confluence, remove the medium, and wash once with sterile PBS.
3. Add 3 mL of 0.25% Trypsin-EDTA to the flask and incubate for ~5 minutes at 37°C. Shake and check every minute until cells detach from the bottom of the plate. Neutralize the enzyme by adding 2-3 volumes of growth medium.
4. Centrifuge 1,000 rpm (~220 g) for 5 minutes and resuspend the cells in desired volume of medium.
5. Seed the cells into new culture vessels with 1:4 dilution.
6. Change the medium every other day until the cells reach 80-90% confluence.

TGFb1 Treatment

1. Seed HHSCs into desired culture vessels with Stellate Cell Growth Medium (Cat # MD-0014).
2. After HHSCs attached (usually in 24 hours), serum starve cells for 12-24 hours using DMEM basal medium without FBS.
3. Treat cells with 2ng TGFb1 in DMEM basal medium for 12-24 hours.
4. Wash twice with PBS. The cells will then be ready for downstream analysis.

References

- [1] Chunyue Yin, et al, J Clin Invest. 2013;123(5):1902–1910. Hepatic stellate cells in liver development, regeneration, and cancer.
- [2] Rockey D. C. . Semin Liver Dis 21(3):337-49. (2001) Hepatic blood flow regulation by stellate cells in normal and injured liver.
- [3] Davidson MD. Integr Biol (Camb). 9(8):662-677.(2017) Microengineered cultures containing human hepatic stellate cells and hepatocytes for drug development.

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