



### PRODUCT INFORMATION

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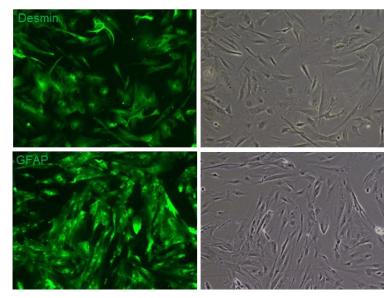
# **Human Hepatic Stellate Cells- Adult (HHSC)**

Catalog Number	10HU-210	Cell Number	1.0 million cells/vial
Species	Homo sapiens	Storage Temperature	Liquid nitrogen

## **Description**

Hepatic stellate cells (HSC) are liver-specific mesenchymal cells, and account for 5~8% of the cells in the liver. HSC play vital roles in the homeostasis of liver extracellular matrix, repair, regeneration and fibrosis, and control retinol metabolism, storage, and release. Stellate cell is the major cell type involved in liver fibrosis in response to liver injury. In healthy liver, HSC 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, HSC 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 HSC is also responsible for secreting collagen scar tissue, which can lead to cirrhosis. In chronic liver disease, prolonged and repeated activation of stellate cells causes liver fibrosis [1,2]. Primary culture of HSC is a valuable tool to study liver fibrosis.

Human Hepatic Stellate Cells (HHSC) from iXCells Biotechnologies are isolated from adult human liver and cryopreserved with ≥ 1 million cells in each vial. HHSC are characterized by immunofluorescence with antibodies specific to Desmin and GFAP. They are negative for HIV-1, HBV, HCV, mycoplasma, bacteria, yeast, and fungi. HHSC can be further expanded for 2-3 passages in iXCells' Stellate Cell Growth Medium (Cat # MD-0014).



**Figure 1.** Human hepatic stellate cells (HHSC) stained with Desmin and GFAP (green). Corresponding phase contrast images of each staining is shown to the right.

#### **Product Details**

Tissue	Adult human liver	
Package Size	1.0 million cells/vial	
Shipped	Cryopreserved	
Storage	Liquid nitrogen	
<b>Growth Properties</b>	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 1-2 minutes until just a tiny ice crystal is left in the cryovial. Keep the cap out of water to minimize the risk of contamination.
- 3. Pipette the cells into a 15 mL conical tube with 5 mL fresh Stellate Cell Growth Medium (Cat # MD-0014).
- **4.** Centrifuge at 1,000 rpm (~220 g) for 5 minutes under room temperature.
- 5. Remove the supernatant and resuspend the cells in desired volume of Stellate Cell Growth Medium.
- It is recommended to seed one vial of HHSC into four 100 mm cell culture dishes using Stellate Cell Growth Medium (Cat # MD-0014).

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

7. 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.

#### Standard Culture Procedure

- 1. When cells reach ~80-90% confluence, remove the medium, and wash once with sterile PBS.
- 2. Add 3 mL of 0.25% Trypsin-EDTA to the cell culture dish 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.
- 3. Centrifuge at 1,000 rpm (~220 g) for 5 minutes and resuspend the cells in desired volume of medium.
- 4. Seed the cells into new culture vessels at 1:4 split ratio.
- 5. Change the medium every other day until the cells reach 80-90% confluence.

#### **TGFβ1 Treatment**

1. Seed HHSC into desired culture vessels with Stellate Cell Growth Medium (Cat # MD-0014).

- 2. After HHSC are attached (usually in 24 hours), serum starve cells for 12-24 hours using DMEM basal medium without FBS.
- 3. Treat cells with 2 ng/mL TGFβ1 in DMEM basal medium for 12-24 hours.
- 4. Wash twice with PBS. The cells will then be ready for downstream analysis.

### References

[1] Yin, C., Evason, K. J., Asahina, K., & Stainier, D. Y. (2013). Hepatic stellate cells in liver development, regeneration, and cancer. The Journal of clinical investigation, 123(5), 1902–1910.

[2] Rockey D. C. (2001). Hepatic blood flow regulation by stellate cells in normal and injured liver. Seminars in liver disease, 21(3), 337–349.

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