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Human Ballooned Hepatocytes: *In Vitro*

Martin Stefanov*

Department of Medical Sciences, Alzaeim Alazhari University, Khartoum North, Sudan

Description

Ballooned Hepatocytes (BH) are aberrant, inflated hepatocytes that are commonly found in liver disorders, particularly non-alcoholic steatohepatitis (NASH). *In vitro* creation of BHs, on the other hand, has proven rare. In this study, primary human hepatocytes were co-cultured with normal human dermal fibroblasts in a cell sheet-based Three-dimensional (3D) model to create human BHs *in vitro*. After only a few days of cultivation, hepatocyte enlargement (2.3 times larger than usual, p 0.01), loss of cytoplasmic keratin, emergence of Mallory-Denk bodies (MDBs), and significant fat droplet formation were seen. In addition, the 3D model revealed ultrastructural features of BHs in human NASH, such as larger mitochondria with crystalline inclusions, dilated endoplasmic reticulum, and MDB development. Furthermore, pathologic aspects of human NASH were discovered, such as enhanced sonic hedgehog ligand production and myofibroblast activation [1].

Human ballooning hepatocytes (BH) seen in non-alcoholic steatohepatitis (NASH) are mostly researched using human liver biopsies and animal models. Human BHs can be successfully replicated in a cell sheet-based *in vitro* model in this study, which is the first *in vitro* model to recapitulate so many histological and ultrastructural characteristics of BHs identified in human NASH, as far as we know. In addition, certain NASH pathophysiological traits were discovered in this investigation. This model could help researchers better understand hepatocellular ballooning and aid translational preclinical medication development in NASH [1, 2].

Non-alcoholic Fatty Liver Disease (NAFLD) is defined by fat deposition in the liver (hepatic steatosis) without a history of excessive alcohol consumption. It is one of the most frequent chronic liver disorders in developed countries. As a progressive form of NAFLD, non-alcoholic steatohepatitis (NASH) can cause liver fibrosis, which can develop to cirrhosis, liver cancer, and eventually liver failure. Despite the fact that NASH is becoming more common around the world, there are still no recognised treatments. This is most likely due to a lack of knowledge about disease mechanisms and appropriate experimental models. The traditional approach for diagnosing NASH has been a liver biopsy. Hepatocellular ballooning is an important histologic characteristic in the diagnosis of NASH [3].

In most cases, ballooning hepatocytes (BH) are described as swollen cells with fat droplets, pale, flocculent cytoplasm, and Mallory-Denk bodies (MDB). The lack of immunostaining for cytokeratin 8/18 has been discovered to be an objective sign of hepatocellular ballooning. Increased sonic hedgehog (SHH) ligand production is another significant property of BHs. The SHH pathway is necessary for liver regeneration and repair, but prolonged activation causes liver fibrosis. Hepatocellular expansion, fat droplet accumulation, dilated endoplasmic reticulum, and expanded mitochondria and MDBs are all ultrastructural characteristics of hepatocellular ballooning in NASH [4].

***Address for Correspondence:** Martin Stefanov, Department of Medical Sciences, Alzaeim Alazhari University, Khartoum North, Sudan, E-mail: martinstef@gmail.com*

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Immunohistochemistry staining

Cell sheet samples were fixed in 4 percent paraformaldehyde at room temperature for 1 hour at predefined time intervals. Standard histological staining with hematoxylin and eosin was performed on fixed cell sheets embedded in paraffin, cut into 4 m slices, and deparaffinized (HE). Sections were processed with Dako proteinase K for antigen retrieval, incubated in Dako REAL peroxidase-blocking solution (Agilent) to quench endogenous peroxidase activity, and then blocked in Blocking One Histo for immunostaining of cytokeratin 8/18 (CK8/18). After that, the sections were incubated with mouse anti-human CK8/18 at a 25-fold dilution, followed by donkey anti-mouse IgG H&L conjugated with horseradish peroxidase (Abcam). The nuclei were stained with hematoxylin and the slices were dyed using Dako Liquid DAB+ Substrate Chromogen System (Agilent).

Immunofluorescence staining

The following primary antibodies were used to incubate Paraffin-embedded sections: CK8/18 or E-Cadherin/-smooth muscle actin (-SMA) or E-Cadherin/-smooth muscle actin (-SMA). Secondary antibodies utilised for staining included Alexa Fluor 488 goat anti-rabbit and/or Alexa Fluor 594 goat anti-mouse. 4′,6-diamidino-2-phenylindole was used to stain the nuclei of cells (DAPI). Finally, the sections were mounted, dried, and photographed using either a confocal laser scanning microscope (CLSM) or a fluorescence microscope (Olympus, Tokyo, Japan) (Nikon). PHHs with positive DAPI staining on the E-Cadherin/DAPI staining images were chosen for quantification of cross-section area per PHH using Image J software. Three cell sheet samples were used for measurement. More than 25 cells were counted in each sample [5].

Cirrhosis is frequently preceded by hepatocyte damage, although this is inadequate for its progression. The liver can regenerate to a normal architecture without persistent fibrosis even after severe hepatocyte necrosis. Furthermore, despite the fact that considerable hepatocyte damage occurs throughout the hepatic lobule in chronic hepatitis, fibrous septa only form in a few areas. Some fibrous septa form as scar tissue to compensate for bridging necrosis induced by direct hepatocyte injury. Some septa, on the other hand, arise as a result of parenchymal extinction following damage and thrombosis of intrahepatic arteries and veins, resulting in parenchymal atrophy. This harm is most likely a 'bystander' effect of the inflammatory response directed at, but not limited to, the infected parenchymal cell in chronic hepatitis.

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Conflict of Interest

The author shows no conflict of interest towards this manuscript.

References

1. Hasui, Nobuhiro, Katsuhisa Sakaguchi, Tetsuya Ogawa,and Yoshihiro Sakamoto, et al. "In vitro ballooned hepatocytes can be produced by primary human hepatocytes and hepatic stellate cell sheets." *Sci Rep* 12, (2022): 1-11.
2. Imashiro, Chikahiro, Kai Yamasaki, Ryu-ichiro Tanaka, and Yusuke Tobe, et al. "Perfusable system using porous collagen gel scaffold actively provides fresh culture media to a cultured 3d tissue." *Int J Mol Sci* 22 (2021): 6780.
3. Gao, Botao, Katsuhisa Sakaguchi, Tetsuya Ogawa, and Yuki Kagawa, et al.

"Functional Analysis of Induced Human Ballooned Hepatocytes in a Cell Sheet-Based Three Dimensional Model." *J Tissue Eng Regen Med* 18 (2021): 217-224.

4. Imashiro, Chikahiro, and Tatsuya Shimizu. "Fundamental technologies and recent advances of cell-sheet-based tissue engineering." *Int J Mol Sci* 22 (2021): 425.

5. Sakai, Yusuke, Makiko Koike, Daisuke Kawahara,and Hideko Hasegawa, et al. "Controlled cell morphology and liver-specific function of engineered primary hepatocytes by fibroblast layer cell densities." *J Biosci Bioeng* 126 (2018): 249-257.

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