Human Hepatocyte/Kupffer cell 3D Spheroid Co-cultures: Characterization and Application for DILI Studies

Patrina Gunnnes,1 Albert P. Li2
1In Vitro ADMET Laboratories, Malden, MA; 2In Vitro ADMET Laboratories, Columbia, MD

Introduction

• Drug-induced liver injury (DILI) is an unanticipated clinical event that can lead to acute liver failure, and results in the termination of drug development program and the application of regulatory restrictions, including boxed warnings.
• The well documented limitations of current 2D in vitro hepatocyte cell culture and in vivo animal models does not allow for the accurate prediction of DILI in humans. There is an urgent need for improved in vitro human hepatocyte models to address these limitations and allow for more accurate prediction of DILI in humans.
• The 3D cell culture of hepatocytes is a rapidly expanding field in an attempt to recreate, in a controlled, artificial environment; the complex 3D microenvironment of the human liver; which is essential for hepatocyte longevity and function. A hepatocyte 3D spheroid culture is a 3D cell culture of aggregated, aggregates of hepatocytes.
• The goal of the study was to develop a procedure to reproducibly culture human hepatocytes and Kupffer cells as spheroids and to characterize these long-term cultures for their suitability for DILI studies.
• Results show that these long-term spheroid cultures (1) have liver-like morphology and function, (2) are sensitive to prototypical hepatotoxicants, (3) respond to an inflammatory stimulus and (4) can distinguish between hepatotoxic and non hepatotoxic compounds.
• Taken all together, the data suggest that these long-term spheroid cultures may be a useful tool to study DILI.

Materials & Methods

Culture of human hepatocyte spheroids:

• Spheroids ready for use
• Albumin production
• IL-6 production

Results

Prototypical hepatotoxicants induce cytotoxicity in human hepatocyte spheroids

Acetaminophen
Letrozole
Triglafenase

Human hepatocyte spheroids can distinguish between a hepatotoxic and non-hepatotoxic compound

Conclusions

• Human hepatocyte spheroid cultures have long-term liver-like morphology & functionality.
• Human hepatocyte/Kupffer cell spheroid co-cultures respond to an inflammatory stimulus, suggesting that it may be useful tool for inflammation-mediated toxicity.
• Prototypical hepatotoxicants induce cytotoxicity in human hepatocyte spheroid cultures.
• Exposure to an inflammatory stimulus increases the sensitivity of hepatocyte/Kupffer cell spheroid co-cultures to trovafloxacin exposure.
• Human hepatocyte spheroid cultures can distinguish between a hepatotoxic and a non-hepatotoxic compound.
• Taken all together, the data suggest that these long-term 3D hepatocyte/Kupffer cell spheroid cultures may be a useful tool for preclinical drug metabolism and toxicity studies, including inflammation-mediated hepatotoxicity.

*Corresponding author: Albert Li (lialbert1@invitroadmet.com)