Cryopreserved HepaRG™ Cells: An Alternative In Vitro Screening Tool for Human Hepatic Drug Metabolism, Induction of Metabolism, & Safety Applications

Jonathan P. Jackson, Manda Edwards, Erica Deibert, and Stephen S. Ferguson, Life Technologies, Cell System Division, ADME/Tox Business Unit, 4301 Emperor Blvd., Durham, NC 27703

ABSTRACT/INTRODUCTION—Currently, primary human hepatocytes (PHHs) are used as the ‘gold standard’ in vitro hepatic model system due to their ability to support mature hepatic phenotypes (e.g. metabolism, transport, and induction) important in drug development, drug-drug interaction (DDI), and safety assessments. However, the use of PHHs for screening applications to identify potential DDI or safety liabilities has been limited by availability, lot-to-lot variability, finite lifespan, and cost. Therefore, significant efforts have been made to develop alternative in vitro systems that can approximate these characteristics of PHHs. For instance, HepaRG™ Cells, human hepatocyte-like cells derived from a single hepatocyte, show comparable functionality to PHH up to 22 days. We also show that cryopreserved HepaRG™ Cells maintain CAR responsiveness with specific substrate-dependent enzyme activity, comparable to PHH. These results coupled with demonstrating the longevity (≥22 days) of HepaRG™ cells in culture highlight hepatic model system’s potential in the study and prediction of xenobiotics clearance, DDI, and safety assessment applications.

RESULTS—Figure 1. Morphology

Table 1. Baseline DME Activity

Table 2. Inter-Lot Reproducibility

Figure 4. Inter-Lot Reproducibility

Figure 5. Induction of P450 Activity

Figure 6. P450 Enzyme Induction Regulatory Pathways

Figure 7. Bile Canaliculi Formation

CONCLUSIONS—

- Baseline P450 Activities in HepaRG™ Cells were comparable to those observed in PHH preparations.
- HepaRG™ Cells support phase I and phase II enzyme expression and function and demonstrate comparable functionality and drug metabolizing enzymes for ≥22 days in culture.
- CYP450 activity induction responses in HepaRG™ Cells were consistent with the induction responses observed in PHH preparations treated with the prototypical hepatic inducers of xenobiotic metabolism.
- Data demonstrated that all major P450 enzyme regulatory pathways (CAR, PXR, and AhR) were functional in HepaRG™ Cells, unlike HepoG2 and Fa2N-4 Cells that lack liver-like CAR expression (6).
- HepaRG™ Cells support bile canalicus formation and the accumulation of CDF within these structures suggests that functional efflux transporters (e.g. MRP2) are present in HepaRG™ cells.
- Uptake activity studies demonstrate that functional uptake transporters (e.g. NTCP and OATP) are present in HepaRG™ cells.
- HepaRG™ Cells support metabolism-dependent toxicity mechanisms.

REFERENCES—

5. Sallusti et al. (2000) CB 166:70-73

Material and Methods Continued—

CYP2C9 Genotype in HepaRG = *2/*2
** CYP2D6 Genotype in HepaRG = *2/WT and *9/WT

For each result only. Not intended for human or animal therapeutic or diagnostic use.

© 2011 Life Technologies Corporation. All rights reserved. The trademarks mentioned herein are the property of Life Technologies Corporation or their respective owners. Thad™ is a registered trademark of Roche Molecular Systems, Inc. HepaRG™ is a trademark of SAB BioPredic International. CHRM is a registered trademark of Advanced PhCsys® Inc.