HEPAC²: Serum-free, Standardized and Validated (Re-usable) Primary Human Hepatocytes for the Analysis of Xenobiotics

Dieter Runge and Anett Ullrich

PRIMACYT Cell Culture Technology GmbH, Hagenower Str. 73, D-19061 Schwerin, www.primacyt.de

Hepatocytes are used as biosensors for pharmacological-toxicological assays to monitor the metabolism and the influence of xenobiotics on hepatocellular functions. We established HEPAC², a long-term culture system in which hepatocytes are plated and cultured under serum-free conditions for up to 3-4 weeks. The hepatocytes retain their specific functions determined by the quantification of urea and albumin release and stay vital.

Due to the maintenance of functionality, the same hepatocytes can be used more than once for assaying the effects of xenobiotics on hepatocellular functions, viability and metabolism studies, since the readout is done non-invasively and the hepatocytes remain intact.

We used Acetaminophen to demonstrate the feasibility of our culture system. The drug-mediated effects did not even change between the first cycle of application on day 4 and the fourth cycle started on day 20 (Ullrich et al. 2007).

In addition, this model was also used successfully to screen the toxic potential of the anticancer drug cis-4-hydroxy-proline. The safety of the drug was documented and in line with the clinical data from a human study, while the data generated in animals were not consistent with the clinical data and let to false conclusions (Dickens et al. 2008).

The robustness of HEPAC² is also shown by repetitive inductions of CYP450 activities. Prototypical inducers increased the enzyme activities to identical levels no matter if induction is started on day 3, day 7 or day 11 of culture.

However, at present, there is no regulatory acceptance for this model. The method has to be tested with further model substances and in external laboratories in order to become an accepted method.