The detection of cholestasis-inducing agents in cultured primary rat hepatocytes

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SCOPE OF THE METHOD

The Method relates to	Human health
The Method is situated in	Basic Research, Translational - Applied Research
Type of method	In vitro - Ex vivo
This method makes use of	Animal derived cells / tissues / organs
Species from which cells/tissues/organs are derived	Rat
Type of cells/tissues/organs	Primary rat hepatocytes

DESCRIPTION

Method keywords

Sandwich cultures

Hepatocytes
Bile salt export pump (Bsep) inhibition
Cholyl-lysyl-fluorescein (CLF)
Cholestasis-inducing potential

Scientific area keywords

Toxicology
in vitro
Drug-induced liver injury (DILI)
cholestasis

Method description

The standard operating procedure describes a method to assess the cholestasis-inducing potential of chemicals, in casu in cultures of primary rat hepatocytes. The procedure relies on the accumulation of the fluorescent bile salt export pump (Bsep) substrate cholyl-lysyl-fluorescein (CLF) in the canalicular network of sandwich-cultured rat hepatocytes either in presence or the absence of Bsep inhibitors.

Lab equipment

Fluorescent microscope (Nikon Eclipse Ti-S, Belgium)

PROS, CONS & FUTURE POTENTIAL

Advantages

The standard operating procedure comprises an easy-to-apply method to detect cholestasis-inducing agents based on Bsep inhibition. Since sandwich cultures of hepatocytes, in contrast to conventional monolayer cultures, exhibit reformation of the canalicular network and polarized excretory functions, this culture systems forms

an appropriate experimental setting for studying biliary excretion.

Challenges

Most Bsep substrates, including CLF, cannot undergo efficient cellular translocation without the support of an uptake transporter, such as sodium-dependent taurocholate cotransporting polypeptide (Ntcp). A number of drugs, known to inhibit Bsep activity, also possess the ability to interfere with the Ntcp-mediated uptake of bile salts. This phenomenon should always be taken into account as it may complicate the interpretation of the experimental results.

REFERENCES, ASSOCIATED DOCUMENTS AND OTHER INFORMATION

References

Dawson, S., Stahl, S., Paul, N., Barber, J. & Kenna, J. G. 2012. In vitro inhibition of the bile salt export pump correlates with risk of cholestatic drug-induced liver injury in humans. Drug Metab Dispos, 40, 130-8

Kaplowtiz, N. 2004. Drug-induced liver injury. Clin Infect Dis, 38 Suppl 2, S44-8

Kis, E., Ioja, E., Rajnai, Z., Jani, M., Méhn, D., Herédi-Szab□, K. & Krajcsi, P. 2011. BSEP inhibition - In vitro screens to assess cholestatic potential of drugs. Toxicol In Vitro

Padda, M. S., Sanchez, M., Akhtar, A. J. & Boyer, J. L. 2011. Drug-induced cholestasis. Hepatology, 53, 1377-87

Schuster, D., Laggner, C. & Langer, T. 2005. Why drugs fail - A study on side effects in new chemical entities. Current Pharmaceutical Design, 11, 3545-59

Swift, B., Pfeifer, N. D. & Brouwer, K. L. 2010. Sandwich-cultured hepatocytes: an in vitro model to evaluate hepatobiliary transporter-based drug interactions and hepatotoxicity. Drug Metab Rev, 42, 446-71

Vinken, M., Elaut, G., Henkens, T., Papeleu, P., Snykers, S., Vanhaecke, T. & Rogiers, V. 2006. Rat hepatocyte cultures: collagen gel sandwich and immobilization cultures. Methods Mol Biol, 320, 247-54

Associated documents

BSEP inhibition.docx

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