LipidTOX assay in primary rat hepatocytes

Commonly used acronym: *LipidTOX*

Created on: 05-03-2019 - Last modified on: 14-08-2025

Organisation

Name of the organisation Vrije Universiteit Brussel (VUB)
Department Pharmaceutical and Pharmacological Sciences
Specific Research Group or Service In Vitro Toxicology and Dermato-Cosmetology
Country Belgium
Geographical Area Brussels Region

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
Species from which cells/tissues/organs are derived	Rat
Type of cells/tissues/organs	Primary rat hepatocytes

DESCRIPTION

Method keywords

Steatosis
Phospholipidosis
Hepatocytes
neutral lipids
phospholipids
Hepatotoxicity

Scientific area keywords

Hepatotoxicity
cytotoxicity
Steatosis
Phospholipidosis

Method description

The method detects two facets of drug-induced cytotoxicity i.e. the intracellular accumulation of phospholipids and of neutral lipids, i.e. phospholipidosis and steatosis respectively. The assay makes use of a kit containing an aqueous, red-fluorescent formulation of labelled phospholipids (LipidTOX™ Red phospholipid stain, excitation/emission ~595/615 nm) which is up taken by the cells upon incubation with a phospholipidosis-inducing compound. The second component of the kit is a selective green-fluorescent stain for neutral lipids (LipidTOX™ Green neutral lipid stain, excitation/emission ~495/505 nm), which can be used sequentially on fixed cells for the analysis of steatosis or can be used independently for single-parameter analysis (Nioi et al. 2007). Additionally, use of VECTASHIELD® Mounting Medium containing 4',6-diamidino-2-phenylindole (DAPI) which binds directly to DNA and produces upon excitation a blue fluorescence, enables intracellular localisation of the lipids.

Lab equipment

Fluorescence microscope

Method status

History of use

REFERENCES, ASSOCIATED DOCUMENTS AND OTHER INFORMATION

References

Anderson N. and Borlak J. (2006) "Drug-induced phospholipidosis." FEBS Letters 580: 5533-40

Elaut G., Henkens T., Papeleu P., Snykers S., Vinken M., Vanhaecke T. and V. Rogiers (2006) "Molecular mechanisms underlying the dedifferentiation process of isolated hepatocytes and their cultures." Current Drug Metabolism 7: 629-60

Gomez-Lechon M.J., Lahoz A., Gombau L., Castell J.V. and Donato M.T. (2010) "In vitro evaluation of potential hepatotoxicity induced by drugs." Current Pharmaceutical Design 16: 1963-77

Kodavanti U.P. and Mehendale H.M. (1990) "Cationic amphiphilic drugs and phospholipid storage disorder." Pharmacological Reviews 42:327-54

McKim J.M., (2010) "Building a tiered approach to in vitro predictive toxicity screening: a focus on assays with in vivo relevance." Combinatorial Chemistry and High Throughput Screening 13: 188-206

Monteith D.K., Morgan R.E. and Halstead B. (2006) "In vitro assays and biomarkers for drug-induced phospholipidosis." Expert Opinion on Drug Metabolism and Toxicology 2: 687-96

Morgan R.E., Kriauciunas A., Berridge B., Sullivan J. and Monteith D.K. (2004) "An in vitro surrogate for drug-induced phospholipidosis." Toxicologist 78: 388

Nioi P., Perry B.K.,. Wang E.J, Gu Y.Z. and R.D. Snyder (2007) "In vitro detection of drug-induced phospholipidosis using gene expression and fluorescent phospholipid based methodologies." Toxicological Sciences 99: 162-73

Papeleu P., Vanhaecke T., Henkens T., Elaut G., Vinken M., Snykers S. and Rogiers V. (2006) "Isolation of rat hepatocytes." Methods in Molecular Biology 320: 229-37

Reasor M.J., Hastings K.L. and Ulrich R.G. (2006) "Drug-induced phospholipidosis: issues and future directions." Expert Opinion on Drug Safety 5:567-83

Sawada H., Takami K. and Asahi S. (2005) "A toxicogenomic approach to drug-induced phospholipidosis: analysis of its induction mechanism and establishment of a novel in vitro screening system." Toxicological Sciences 83:282-92

Sawada H., Taniguchi K. and Takami K. (2006) "Improved toxicogenomic screening for drug-induced phospholipidosis using a multiplexed quantitative gene expression ArrayPlate assay." Toxicology In Vitro 20:1506-13
Seglen, P.O. (1976) "Preparation of isolated rat liver cells." Methods in Cell Biology 13:29-83

Links

IVTD - VUB
Prof. Mathieu Vinken - Team

Coordinated by

Financed by