Sudan Red III in situ staining of cultured primary rat hepatocytes

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SCOPE OF THE METHOD

Alternative method relates to	Human health
Alternative method is situated in	Basic Research, Translational - Applied Research
Type of alternative 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

Formaldehyde fixation

Sudan Red III staining

Hematoxylin nuclear counterstain

Primary rat hepatocytes

Intracellular lipids

in vitro

Scientific area keywords

Toxicology
Hepatotoxicity
Steatosis
Drug-induced cytotoxicity

Method desription

The standard operating procedure for Sudan Red III in situ staining of cultured rat hepatocytes describes how to detect one of the aspects of drug-induced cytotoxicity i.e. the intracellular accumulation of lipids or in other words steatosis, in primary rat hepatocyte cultures. It is based on the ability of a lysochrome, i.e. Sudan Red III diazo-dye to stain intracellular lipids. Additionally, subsequent application of hemalum, which is a complex formed by aluminium ions and oxidized haematoxylin, colours nuclei of the cells and thus enables their localisation. Red-coloured lipid droplets and blue nuclei are readily visible upon examination of the cells under a light microscope.

Lab equipment

Inverse-phase light microscope (Nikon Optiphot)
Oven (Thermo electron corporation, Heraeus, 60°C)

PROS, CONS & FUTURE POTENTIAL

Advantages

The standard operating procedure for Sudan Red III in situ staining of cultured primary rat hepatocytes is easily applicable and allows a simultaneous screening of multiple compounds and/or multiple concentrations of the same compounds (to examine chemically induced steatosis).

Challenges

Sudan Red III stain has a high affinity to a broad range of lipids and consequently does not discriminate between e.g. neutral lipids and phospholipids. Therefore, it is of utmost importance to perform more than one assay or use a more specific assay.

REFERENCES, ASSOCIATED DOCUMENTS AND OTHER INFORMATION

Associated documents

Sudan Red III staining.doc

PARTNERS AND COLLABORATIONS

Organisation

Name of the organisation Vrije Universiteit Brussel

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Country Belgium

Coordinated by

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