

# Measurement of the extracellular release of lactate dehydrogenase in cultured primary rat hepatocytes

Commonly used acronym: LDH assay

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# 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
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

Hepatotoxicity
Hepatocytes
cytotoxicity
LDH

# Scientific area keywords

Toxicology
Hepatotoxicity
Primary hepatocytes
cytotoxicity

# **Method description**

This method assesses general cytotoxicity. Upon disruption of the cell membrane, lactate dehydrogenase (LDH) is released. LDH catalyzes the interconversion of pyruvate and lactate with concomitant interconversion of reduced (NADH) and oxidized (NAD+) nicotinamide adenine dinucleotide. The principle of the assay described in the current standard operating procedure is based on this reaction. In particular, the consumption of NADH is spectrophotometrically assessed and serves as a measure that is proportional to the LDH activity.

# Lab equipment

Spectrophotometer

#### **Method status**

History of use

# PROS, CONS & FUTURE POTENTIAL

#### **Advantages**

Easy-to-apply method

#### **Challenges**

Cell membrane damage is a rather late and rough marker of cytotoxicity that mainly indicates necrosis and that may yield false negative results

### REFERENCES, ASSOCIATED DOCUMENTS AND OTHER INFORMATION

#### References

Bergmeyer H.U. (1974) Lactate dehydrogenase. In: Methods of enzymatic analysis, (ed. H.U. Bergmeyer), pp. 574-579. New York, USA: Academic Press Elaut G., Henkens T., Papeleu P., Snykers S., Vinken M., Vanhaecke T. and Rogiers V. (2006) Molecular mechanisms underlying the dedifferentiation process of isolated hepatocytes and their cultures. Current Drug Metabolism 7: 629-660 Gómez-Lechón 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: 1963-1977

Jaeschke H., Gores G.J., Cederbaum A.I., Hinson J.A. Pessayre D. and Lemasters J.J. (2002) Mechanisms of hepatotoxicity. Toxicological Sciences 65: 166-176 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

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: 229-237 Seglen P.O. (1976) Preparation of isolated rat liver cells. Methods in Cell Biology 13: 29-83

Van Calsteren V. (2010) Analyse van de herhaalde toedieningstoxiciteitstesten met cosmetische ingrediënten bestudeerd op Europees niveau. Dissertation Master in Pharmaceutical Sciences - Vrije Universiteit Brussel, Rogiers V. (promotor) and Pauwels M. (co-promotor)

Vinken M., Decrock E., De Vuyst E., Leybaert L., Vanhaecke T. and Rogiers V. (2009) Biochemical characterisation of an in vitro model of hepatocellular apoptotic cell death. Alternatives to Laboratory Animals 37: 209-218

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