

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

**Commonly used acronym:** LDH assay

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### Contact person

Kaat Leroy

### 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

<b>The Method relates to</b>	Human health
<b>The Method is situated in</b>	Basic Research
<b>Type of method</b>	In vitro - Ex vivo
<b>Species from which cells/tissues/organs are derived</b>	Rat
<b>Type of cells/tissues/organs</b>	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<sup>+</sup>) 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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