IN CYTOTOX

Manufactured By: Xenometrix AG

LDHe - GLU - XTT - SRB

APPLICATIONS

Combined colorimetric assays for the quantification of the membrane integrity, cellular and mitochondrial metabolism and total protein synthesis rate of cells in response to pharmaceutical, chemical and environmental compounds, and nutrients.

PRINCIPLE

This kit allows to measure sequentially four cytotoxicity parameters in one single cell culture: Membrane integrity **(LDHe: Extracellular Lactate Dehydrogenase)**, metabolic activity **(GLU: Glucose)**, mitochondrial activity **(XTT: Tetrazolium Hydroxide)** and total protein synthesis rate **(SRB: Sulforhodamine B)**.

Released LDH, a marker for cell damage, is determined kinetically in the medium (NADH consumption). Extracellular glucose concentration (GOD / POD method) is inversely proportional to the rate of glucose uptake, which is an indicator of the functional metabolic state of cells. XTT is reduced in the cells to formazan by mitochondrial succinate dehydrogenase. The reduction rate is measured and correlates with mitochondrial activity. The fixed SRB dye, measured photometrically after solubilization, correlates with total protein synthesis rate and therefore with cell proliferation.

Unlike some other LDHe assays, the In Cytotox LDHe assay measures the oxidation of NADH to NAD+ and the concurrent reduction of pyruvate to lactate. By providing an excess of pyruvate in the reaction mixture, the In Cytotox LDHe assay is therefore insensitive to pyruvate in the culture medium, which can cause product inhibition of the reverse reaction implemented in other LDHe assays.

BIOLOGICAL PARAMETERS EVALUATION

- IC_{50} (Inhibitory Concentration 50%)
- Membrane integrity
- Glucose consumption rate
- Respiratory chain activity
- Total protein synthesis
- Cell proliferation

TECHNICAL SPECIFICATIONS

Absorbance:

- LDHe: 340 nm
- GLU: 540 nm
- XTT: 480 (optimum) or 450 nm
- SRB: 540 nm
  (Recommended reference filter: 690 nm)

Approximate assay time (total):

- 6 hr 40 + time to dry the plate (SRB) if all steps are done sequentially. Some incubations may be done in parallel (LDHe, Glu).

Available kit configurations:

- Reagents only
- Reagents and 96-well microplates, sterile reagent reservoirs

LDHe-GLU-XTT-SRB Kit content:

- Buffer solutions
- Enzyme solution
- Stop solution
- Substrate solutions
- Activator solution
- Labeling solution
- Wash solution
- Solubilization solution
- Rinsing solution
- Fixing solution
- Instruction manual

LDHe GLU XTT SRB KIT SIZES AND REFERENCE NUMBERS

REFERENCE NO.	NUMBER OF TESTS
AKLGXS96.300	4 x 300
AKLGXS96.1200	4 x 1200

Kits with plasticware (microplates and reservoirs) are also available.
Individual reagents and other kit sizes available upon request.

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