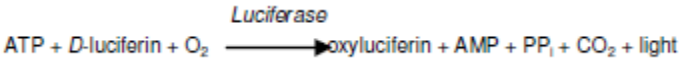
Cytotoxicity Assay Kit (Chemiluminescent)

LS-K256-100 (100 Tests) • **Store at -20°C**

LSBio
LifeSpan BioSciences, Inc.

Introduction

Adenosine 5'-triphosphate (ATP) is the chemical energy for cellular metabolism and is often referred to as energy currency of the cell. ATP is produced only in living cells during photosynthesis and cellular respiration and consumed in cellular processes including biosynthetic reactions, motility and cell division. It is a key indicator of cellular activity and has been utilized as a measure of cell viability and cytotoxicity in research and drug discovery. This Cytotoxicity Assay Kit provides a rapid Method to measure intracellular ATP, cell viability and cytotoxicity. The single working reagent lyses cells to release ATP, which, in the presence of luciferase, immediately reacts with the Substrate *D*-luciferin to produce light. The light intensity is a direct measure of intracellular ATP concentration and hence number of living cells.



This non-radioactive, homogeneous cell-based assay can be conveniently performed in microplates. The reagent is compatible with all culture media and liquid handling systems for high-throughput screening applications in 96-well and 384-well plates.

Key Features

- Safe. Non-radioactive assay (cf. ^{3}H-thymidine incorporation assay).
- Sensitive and accurate. As low as 50 cells can be quantified.
- Homogeneous and convenient. "Mix-incubate-measure" type assay. No wash and reagent transfer steps are involved.
- Robust and amenable to HTS: Z' factors of 0.6 to 0.7 are routinely observed in 96-well and 384-well plates. Can be readily automated on HTS liquid handling systems for processing thousands of samples per day.

Applications

- Cell proliferation: effects of cytokines, growth factor, nutrients.
- Cytotoxicity and apoptosis: evaluation of toxic compounds, anti-cancer antibodies, toxins, environmental pollutants, etc.
- Drug discovery: high-throughput screening for anticancer drugs.

Components

Component	K256-100
	100 Tests
Assay Buffer	10 mL
Substrate	120 µL
ATP Enzyme	120 µL

Storage

The kit is shipped on ice. Store all reagents at -20°C. Shelf life: 6 months after receipt.

Cytotoxicity Assay Kit (Chemiluminescent)

LS-K256-100 (100 Tests) • **Store at -20°C**

Assay Procedure

Assay Procedure in 96-Well Plates

1. Cell Culture. Plate cells at 100 µL/well in white opaque tissue culture plates. If desired, add 5 µL test compounds and controls dissolved in PBS or culture medium per well. Rock plate lightly to mix and incubate for desired period of time (e.g. overnight).
2. Assay. Bring all components to room temperature. Keep thawed ATP Enzyme on ice or 4°C. For each test well, mix 95 µL Assay Buffer with 1 µL Substrate and 1 µL ATP Enzyme. Add 90 µL Reconstituted Reagent to each test well and mix by tapping the plate. Incubate for 2 minutes at room temperature.

Read luminescence on a luminometer. For most luminometers (Berthold Luminometer, LJL Analyst, Top Count, MicroBeta Counters, CLIPR and LeadSeeker), integration time of 0.1 to 5 sec is appropriate.

Assay Procedure in 384-Well Plates

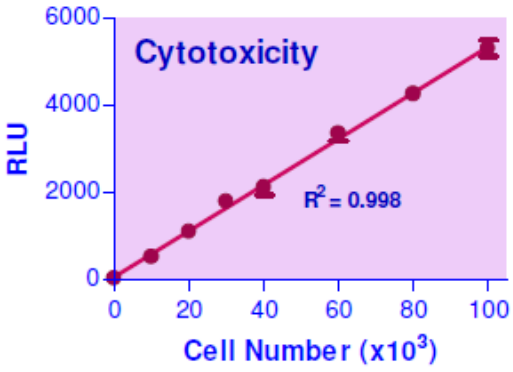
1. Cell Culture. Plate cells at 25 µL/well in white opaque tissue culture plates. If desired, add 5 µL test compounds and controls dissolved in PBS or culture medium per well. Rock plate lightly to mix and incubate for desired period of time (e.g. overnight).
2. Assay. Bring all components to room temperature. Keep thawed ATP enzyme on ice or 4°C. For each test well, mix 30 µL Assay Buffer with 0.3 µL Substrate and 0.3 µL ATP Enzyme. Add 25 µL Reconstituted Reagent to each well and mix by tapping the plate. Incubate for 2 minutes at room temperature.

Read luminescence on a luminometer. For most luminometers (Berthold Luminometer, LJL Analyst, Top Count, MicroBeta Counters, CLIPR and LeadSeeker), integration time of 0.1 to 5 sec is appropriate.

General Considerations

Signal stability. After adding the Reconstituted Reagent, the luminescence signal is stable for about 15 min and decreases slow thereafter. Reading is best performed within 30 min.

Sample Data



Linearity of Luminescence to Cell Number in 96-well Plate Assay

Version: V.08.09.2018

LifeSpan BioSciences, Inc. • 2401 Fourth Avenue, Suite 900, Seattle, WA 98121
www.LSBio.com • (206) 464-1554 • TechnicalSupport@LSBio.com