VKEYBIO-02-2024

For Research Use Only

Not For Diagnostic Or Therapeutic Use

KeyTec® Luminescent Cell Viability Detection Kit

Instruction Manual

1. Introduction

KeyTec® Luminescent Cell Viability Detection kit (LCV) is designed for cell viability detection. It quantifies viable cells by measuring the ATP present, a key indicator of metabolically active cells. The homogeneous assay procedure involves adding a single reagent directly to cells cultured in serum-supplemented medium.

The detection principle is based on Luminescent technology. Within the kit, D-Luciferin and luciferase react with ATP released by cells, generating a sensitive and robust luminescent signal. With a luminescent signal half-life of 3-5 hours, ensuring high sensitivity and robust performance. Additionally, the kit shows excellent storage stability, maintaining above 90% activity after 30 days at room temperature or 60 days at 2-8 °C. Its sensitivity and operational flexibility make it ideal for high-throughput screening (HTS), cell proliferation and cytotoxicity assays (Figure 1).

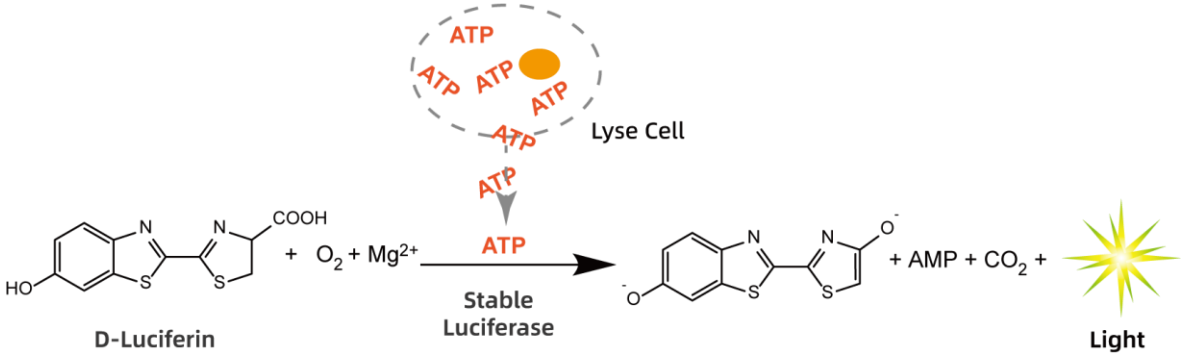

Figure 1. The luciferase reaction of KeyTec® Luminescent Cell Viability detection kit

Shanghai VKEY Biotechnologies Co.,Ltd.
Floor 3, Building 2, Lane 898, Halei Road, Pudong New Area, Shanghai, China
Website：www.vkeybio.com
Email：technical-support@vkeybio.com

2. Components

CAT.	Description	Size
A2010001N	KeyTec® Luminescent Cell Viability Detection Kit (100 tests)	10 mL

Each kit contains sufficient reagents to perform 100 tests of 100 µL/well.

The kit contains the following components:

- 1 × 10 mL KeyTec® Luminescent Cell Viability Detection Reagent

CAT.	Description	Size
A2010002N	KeyTec® Luminescent Cell Viability Detection Kit (1,000 tests)	2*50 mL

Each kit contains sufficient reagents to perform 1,000 tests of 100 µL/well.

The kit contains the following components:

- 2 × 50 mL KeyTec® Luminescent Cell Viability Detection Reagent

CAT.	Description	Size
A2010003N	KeyTec® Luminescent Cell Viability Detection Kit (5,000 tests)	2*250 mL

Each kit contains sufficient reagents to perform 5,000 tests of 100 µL/well.

The kit contains the following components:

- 2 × 250 mL KeyTec® Luminescent Cell Viability Detection Reagent

CAT.	Description	Size
A2010004N	KeyTec® Luminescent Cell Viability Detection Kit (10,000 tests)	4*250 mL

Each kit contains sufficient reagents to perform 10,000 tests of 100 µL/well.

The kit contains the following components:

- 2 × A2010003N KeyTec® Luminescent Cell Viability Detection Kit (5,000 tests)

CAT.	Description	Size
A2010005N	KeyTec® Luminescent Cell Viability Detection Kit (5,000 tests)	10*50 mL

Each kit contains sufficient reagents to perform 5,000 tests of 100 µL/well.

The kit contains the following components:

- 10 × 50 mL KeyTec® Luminescent Cell Viability Detection Reagent

Shanghai VKEY Biotechnologies Co.,Ltd.
Floor 3, Building 2, Lane 898, Halei Road, Pudong New Area, Shanghai, China
Website：www.vkeybio.com
Email：technical-support@vkeybio.com

CAT.	Description	Size
A2010006N	KeyTec® Luminescent Cell Viability Detection Kit (10,000 tests)	20*50 mL

Each kit contains sufficient reagents to perform 10,000 tests of 100 µL/well.

The kit contains the following components:

- 2 × A2010005N KeyTec® Luminescent Cell Viability Detection Kit (5,000 tests)

3. Storage Conditions

- Upon receipt, store the kit below -40 °C. Refer to the COA document for the validity period.
- Once thawed, the reagent can be stored at room temperature (≤ 24.5 °C) for 30 days or at 2-8°C for 60 days (≥90% activity).
- The kit can withstand up to 10 cycles of freezing and thawing (≥90% activity).
- We recommend preparing the mixed reagent immediately before use.

4. Materials Required But Not Supplied

Materials	Recommended Brand	CAT.
Cell Culture Plate (96-well, clear flat bottom, white)	Corning	3610
	Greiner	655098
White Microplates Bottom Seals	VKEY-BIO	M1000302N
Pipettes	Multiple Choices	\
Microplate Shakers	Multiple Choices	\
Microplate Reader With Luminescence	Multiple Choices	\

Shanghai VKEY Biotechnologies Co.,Ltd.
Floor 3, Building 2, Lane 898, Halei Road, Pudong New Area, Shanghai, China
Website：www.vkeybio.com
Email：technical-support@vkeybio.com

5. Assay Procedure

Procedure	Stage	Operation
Step 1	Reagents Preparation	➢ **Thaw the reagents:** Allow the reagent to thaw at 4℃ or room temperature (**not above 25 ℃**) before use. ➢ **Mix reagent after equilibration:** Once the reagent has reached to room temperature, invert the bottle several times to ensure thorough mixing of the reagent. (If a small amount of precipitation occurs, simply mix it several times to dissolve it, and the performance of the reagent is not affected.)
Step 2	Detection	➢ **Equilibrate culture plate temperature:** Equilibrate the cell culture plate to room temperature . ➢ **Add reagent:** Add an equal volume of LCV reagent to the sample to be tested. (It is recommended to add 100 µL of reagent to 100 µL of the cell culture to be tested.) ➢ **Shake the plate:** Shake for 2-5 minutes to achieve thorough cell lysis and mixing, then react at room temperature for 10 minutes to reach maximum luminescent signal. ➢ **Read Signal:** Read the luminescent signal with a microplate reader.

6. Performance

- **Cell density assay data**

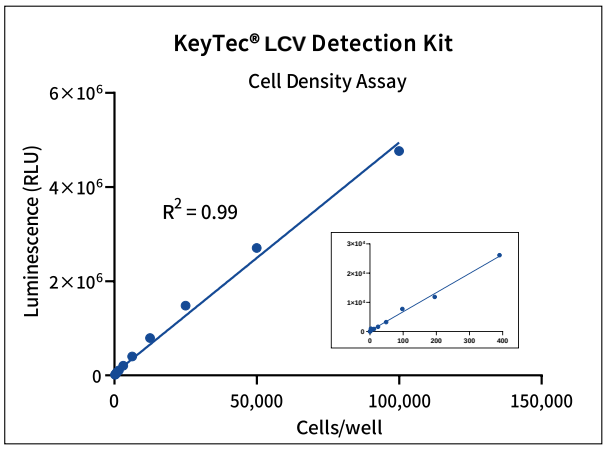

Figure 2. Correlation between Cell Number and Luminescence

Use KeyTec® Luminescent Cell Viability Detection kit to detect cell viability of HeLa cell line. The results showed a linear relationship between the luminescent signal and the linear range across four orders of magnitude. Hela cells, cultured in DMEM medium with 10% FBS, were serially diluted two-fold, starting from 100,000 cells per well in a 96-well plate. The luminescent signal exhibits a linear relationship (R^2 = 0.99) with cell number per well.

Shanghai VKEY Biotechnologies Co.,Ltd.
Floor 3, Building 2, Lane 898, Halei Road, Pudong New Area, Shanghai, China
Website：www.vkeybio.com
Email：technical-support@vkeybio.com

Storage Stability test data

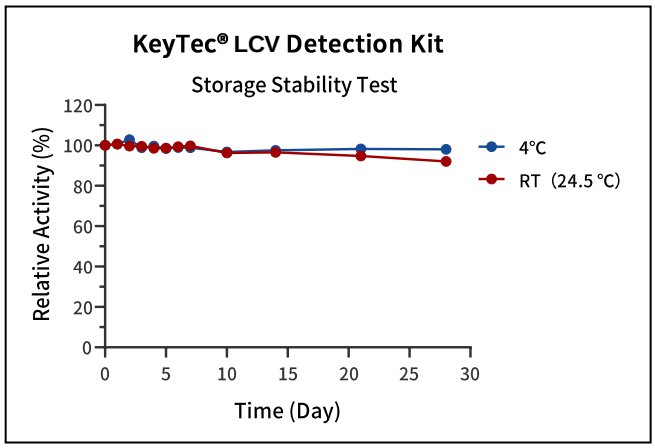

Figure 3. The stability trend of KeyTec® LCV reagent stored at room temperature and 4 °C over time

After incubating equal portions of KeyTec® LCV reagent at 4 °C and 24.5 °C for varying durations, then freeze them at -80°C. Upon thawing to room temperature, mix the reagent samples with an equal volume of 1 μM ATP solution, incubate for 15 minutes, and record luminescence values. Relative activity refers to the luminescence values of samples after different incubation times as a percentage relative to the luminescence value at T = 0.

Tip: The data provided above is for reference only. Actual results may vary depended on the performance of the microplate reader used.

Shanghai VKEY Biotechnologies Co.,Ltd.
Floor 3, Building 2, Lane 898, Halei Road, Pudong New Area, Shanghai, China
Website：www.vkeybio.com
Email：technical-support@vkeybio.com