Bio-UltraBio™ Bright one-step Firefly Luciferase Reporter Gene Assay Kit (Ready-to-use)

B1506502

Storage: -80℃. Protect from light.

Introduction

Genetic reporters are used commonly in cell biology to study gene expression and other cellular events coupled to gene expression, such as receptor activity, intracellular signal transduction, mRNA processing, protein folding and protein: protein interactions. Firefly luciferase is one of the most commonly used reporter genes in gene expression. It is an extremely sensitive, fast-responding and easy-to-use reporter gene. The chemiluminescence reaction it catalyzes is one of the most sensitive analytical tools for measuring gene expression.

Firefly luciferase is a protein with a molecular weight of approximately 61 kDa. In the presence of ATP, magnesium ions and oxygen, it can catalyze the oxidation of luciferin into oxyluciferin. During the oxidation process of luciferin, bioluminescence with a wavelength of approximately 560 nm is emitted, and this fluorescence can be measured by a chemiluminescence analyzer. Bio-UltraBio™ Bright one-step Firefly Luciferase Reporter Gene Assay Kit (Ready-to-use) is a one-step assay kit for directly measuring the intracellular firefly luciferase activity through chemiluminescence without the need for cell washing or collection. It features ultra-high sensitivity and high signal stability. The kit provides ready-to-use firefly luciferase assay reagents. Just add them to the cell culture plate in an equal volume to the culture medium and react for 5 minutes, then chemiluminescence detection can be carried out. This kit is flexible and convenient to use, with high detection sensitivity and relatively stable luminescence signals. The signal half-life is 0.5 to 1 hour.

Kit Contents

B1506502	Component	100 T	10×100 T	Storage conditions	Quantity Per Test
B1506502	Firefly luciferase detection reagent	10 mL	10×10mL	-80℃. Store in the dark.	100 μL

Instructions for Use

1. Cell lysis

1) Adherent cells: Do not aspirate the cell culture medium; directly add the detection reagent of the same volume as the medium.
   Suspension cells: The same as adherent cells.
2) Recommended usage volume:

Culture Vessel	96-well plate	48-well plate	24-well plate	12-well plate	6-well plate
Culture Medium Volume (μl/well)	100	150	200	300	500
Detection Reagent Volume (μl/well)	100	150	200	300	500

3) Lyse the cells for 5 minutes at room temperature, either on a microplate shaker or by standing.

2. Sample Addition

Samples in 96-well plates may proceed directly to the subsequent assay. For other sample types, pipette 200 μl of the mixed solution (culture medium + detection reagent) per well into the assay plate.

3. Detection

Initiate the chemiluminescence detector or a multifunctional microplate reader equipped with chemiluminescence functionality. Set parameters according to the instrument manual and experimental requirements, then proceed to measure the signal values.

Precautions

1. The luciferase detection reagent for fireflies should be avoided from repeated freezing and thawing as much as possible. It is recommended to appropriately aliquot the reagent for the first use and then store it in a dark place at -80°C.
2. The enzymatic reaction is temperature-sensitive. Both the cell culture plate and the detection reagent must be equilibrated to room temperature prior to assay initiation.
3. Detector Selection: Any instrument capable of measuring chemiluminescence is suitable for use with this kit. However, for identical samples, the background signal and measured values may vary between different detectors. Furthermore, readings obtained from different instruments for the same sample are not directly comparable.
4. To prevent well-to-well crosstalk, the use of opaque white or black cell culture plates is recommended.