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17-1101: Leeporter™ Renilla Luciferase Assay Reagent- 1000 Test

Application : Functional Assay

Description

The Leeporter™ Renilla Luciferase Assay Reagent was specifically formulated to use with our Leeporter™ (Luciferase Reporter) Cell Lines expressing an optimized intracellular Renilla luciferase, which was designed to produce highly sensitive signal and prolonged signal intensity. As a one-step glow assay reagent, the Leeporter™ Luciferase Assay Reagent can be directly added to cell culture plates compatible with the luminometer being used without diluting or transferring culture supernatants or cell lysates to other plates.

Components & Storage

Reagent	Amount	Storage
Substrate (100X), lyophilized	1 vial	-20°C
Substrate Reconstitution Solution	0.5 ml	Room temperature or -20°C
Assay Buffer	50 ml	-20°C

Product Info

Amount : 1 kit
Content : 1000 test (96-well plate format). Assay buffer contains 0.01% Sodium Azide.
Storage condition : Store at -20°C

Application Note

Protocol:

1) Substrate (100X) Reconstitution:

Add 0.5 ml Substrate Reconstitution Solution to lyophilized Substrate vial and dissolve thoroughly by gently pipetting up and down, and/or inverting tubes. The reconstituted 100X substrate may be stored in a dark environment at -20°C for up to one month.

2) Assay Buffer:

Thaw Assay Buffer of 50 ml in room temperature water bath and dissolve thoroughly any precipitates by swirling and/or gentle vortexing until solution becomes clear. Assay Buffer can be

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aliquoted (e.g. 5-10 ml each) and stored at -20°C for several months.

3) Preparation of complete Assay Solution (for 96-well plate format):

Prepare complete Assay Solution fresh for each use, which should be used within 2 hours. Thaw Assay Buffer and equilibrate to room temperature with swirling and/or gentle vortexing. Calculate how much complete assay solution is needed (Note: 50 ul of complete assay solution is required for each well of 96-well plate). Aliquot proper amount of Assay Buffer in a 15 ml tube and add corresponding amount of reconstituted 100X substrate to make a final 1X substrate assay solution.

4) Luciferase Assay (96-well plate format):

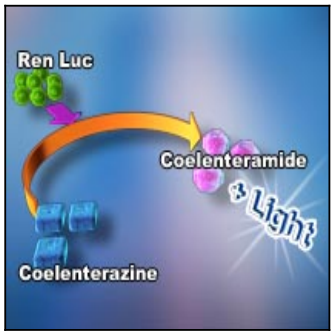
1. Plate you target Leeporter™ Luciferase reporter cells in a white solid-bottom 96-well microplate (Note: The 96-well plate used should be compatible with the luminometer being used.) at 100 ul cells/well, based on the corresponding protocol to your target Leeporter™ cell line (Note: Each target Leeporter™ cell line protocol can be found in its corresponding Data Sheets.). [Total volume per well is 100 uL]

2. Stimulate or treat your target cells based on the corresponding protocol to your target Leeporter™ cell line. [Total volume per well becomes 105~110 uL = 100 ul cells + 5~10 ul treatment]

3. After completion of stimulation/treament of your target cells, equilibrate the 96-well plate containing your target cells being assayed to room temperature for 10 minutes (Note: Do NOT remove or disturb cell culture medium.).

4. Using a multi-channel pipettor, add 50 ul complete Assay Solution directly to each plate well (Note: Do NOT remove cell culture medium when adding the Assay Reagent. So the Assay Reagent of 50 ul should be directly added on top of the cell culture of each well.). [Total volume per well becomes 155~160 uL = 100 ul cells + 5~10 ul treatment + 50 ul complete assay solution]

5. Use automix for 2-3 seconds, and then read the plate in a luminometer within 1-5 minutes. (An example of a luminometer set up: SpectraMaxL (Molecular Devices): target wavelength of 470nm, integration time at 0.5 sec and automix for 2 sec).



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