



Luciferase Assay Reagent

Catalog number: AR4012

This package insert must be read in its entirety before using this product.

For research use only. Not for use in diagnostic procedures.

Catalog Number: AR4012, **Storage:** Store at -20°C for one year. For short-term storage and frequent use, store at 4°C for up to one month. Avoid repeated freeze-thaw cycles.

Overview

Product Name	Luciferase Assay Reagent
SKU/Catalog Number	AR4012
Content	1000 tests (96-well plate format). Assay buffer contains 0.01% Sodium Azide. Sodium Azide is highly toxic.
Description	The Luciferase Assay Reagent was specifically formulated to use with our (Luciferase Reporter) Cell Line products expressing an optimized intracellular Renilla luciferase, which was designed to produce highly sensitive signal and prolonged signal intensity. As one-step glow assay reagent, the 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.

Kit Components

Reagent	Amount	Storage
Substrate (100X), lyophilized	1 vial	-20°C
Substrate Reconstitution Solution	0.5ml	Room Temperature or -20°C
Assay Buffer	50ml	-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 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 your target [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 cell line (Note: Each target cell line protocol can be found in its corresponding datasheets.). [Total volume per well is 100 uL]

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

3. After completion of stimulation/treatment 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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