

QUANTI-Luc™ 4 Renilla

A coelenterazine-based luminescence assay reagent

Catalog code: rep-qlc4r1, rep-qlc4r2, rep-qlc4r5

<https://www.invivogen.com/ quanti-luc>

For research use only

Version 23F28-AK

PRODUCT INFORMATION

Contents

QUANTI-Luc™ 4 Renilla is a two-component reporter kit which contains:

- QUANTI-Luc™ 4 Reagent (20X)
- QUANTI-Luc™ 4 Lysis Buffer (5X)

QUANTI-Luc™ 4 Renilla is supplied in different formats:

- rep-qlc4r1: 1 tube of QUANTI-Luc™ 4 Reagent and 1 bottle of QUANTI-Luc™ 4 Lysis Buffer
- rep-qlc4r2: 2 tubes of QUANTI-Luc™ 4 Reagent and 1 bottle of QUANTI-Luc™ 4 Lysis Buffer
- rep-qlc4r5: 5 tubes of QUANTI-Luc™ 4 Reagent and 2 bottles of QUANTI-Luc™ 4 Lysis Buffer

Each tube of QUANTI-Luc™ 4 Reagent and bottle of QUANTI-Luc™ 4 Lysis Buffer is sufficient for 5 x 96-well plates (25 ml).

Required Material (not provided)

- Sterile water
- Sterile screw cap tube

Storage and Stability

- Store QUANTI-Luc™ 4 Reagent and QUANTI-Luc™ 4 Lysis Buffer at -20°C for up to 12 months.
- After preparation, the QUANTI-Luc™ 4 Reagent working solution is stable for 48 hours at 4°C and 1 month at -20°C. Prepare aliquots to avoid repeated freeze-thaw cycles. After preparation, the QUANTI-Luc™ 4 Lysis Buffer working solution is stable for 1 week at 4°C and 1 month at -20°C. Prepare aliquots to avoid repeated freeze-thaw cycles.

Note: This product is photosensitive and should be protected from light.

Quality Control

Each lot is thoroughly tested to ensure the absence of lot-to-lot variation.

- Physicochemical characterization (pH, appearance).
- Functional assays using Renilla reporter cells.

DESCRIPTION

QUANTI-Luc™ 4 Renilla is an optimized kit for the detection of intracellular **Renilla** activity after cell lysis. This two-component kit comprises QUANTI-Luc™ 4 Reagent (coelenterazine-containing reagent) and QUANTI-Luc™ 4 Lysis Buffer. The light signal produced correlates to the amount of luciferase protein expressed. It is quantified using a luminometer and expressed as relative light units (RLUs).

The pH of QUANTI-Luc™ 4 Reagent allows monitoring the cellular response in small sample numbers or when using microplate readers with reagent injectors.

PREPARATION OF WORKING SOLUTIONS

Below are the instructions to prepare QUANTI-Luc™ 4 Reagent and QUANTI-Luc™ 4 Lysis Buffer working solutions for 5 x 96-well plates. Alternatively, you may prepare the working solutions from the 20X QUANTI-Luc™ 4 Reagent and 5X QUANTI-Luc™ 4 Lysis Buffer to reach the required volume (see summary table on next page).

Preparation of QUANTI-Luc™ 4 Lysis Buffer working solution

1. Dilute the total volume of the 5X bottle (17 ml) of Lysis Buffer into 68 ml sterile water to obtain 85 ml of working solution.
2. Vortex a few seconds.
3. Use the working solution immediately or store until required for use (up to 1 week at 4°C or 1 month at -20°C).

Preparation of QUANTI-Luc™ 4 Reagent working solution

1. Dilute the total volume of the 20X tube (1.25 ml) of Reagent into 23.75 ml sterile water to obtain 25 ml of working solution.
2. Vortex a few seconds.
3. Use the working solution immediately or store until required for use (up to 48 hours at 4°C or 1 month at -20°C).

TECHNICAL SUPPORT

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Volumes of QUANTI-Luc™ 4 Lysis Buffer and Reagent working solutions

| Preparation of Lysis Buffer working solution | | |
|--|---------------------------------|--------------------------|
| Final volume of working solution | QUANTI-Luc™ 4 Lysis Buffer (5X) | Sterile H ₂ O |
| 5 ml | 1 ml | 4 ml |
| 10 ml | 2 ml | 8 ml |
| 25 ml | 5 ml | 20 ml |

| Preparation of Reagent working solution | | |
|---|-----------------------------|--------------------------|
| Final volume of working solution | QUANTI-Luc™ 4 Reagent (20X) | Sterile H ₂ O |
| 5 ml | 0.25 ml | 4.75 ml |
| 10 ml | 0.50 ml | 9.50 ml |
| 25 ml | 1.25 ml | 23.75 ml |

METHODS

Cell lysis

For adherent cells

1. Remove culture supernatant using a pipet or aspiration.
2. Gently rinse cells using 100 µl PBS 1X per well.
3. Remove PBS 1X using a pipet or aspiration.
4. Add 50 µl QUANTI-Luc™ 4 Lysis Buffer 1X working solution per well.
5. Pipet up and down 5 times to lyse the cells and proceed to detection immediately.

For cells in suspension

1. Centrifuge at 150 x g (RCF) for 10 min or 300 x g (RCF) for 5 min.
2. Remove supernatant using a pipet or aspiration.
3. Gently rinse cells using 100 µl PBS 1X per well.
4. Centrifuge at 150 x g (RCF) for 10 min or 300 x g (RCF) for 5 min.
5. Remove PBS 1X using a pipet or aspiration.
6. Add 50 µl QUANTI-Luc™ 4 Lysis Buffer 1X working solution per well.
7. Pipet up and down 5 times to lyse the cells and proceed to detection immediately.

Detection of luciferase activity from cell lysis

Prepare the QUANTI-Luc™ 4 Reagent working solution as per instructions (see section "preparation of working solutions").

To obtain **end-point readings** using a luminometer **with an injector**.

1. Set the luminometer with the following parameters: 50 µl of injection, end-point measurement with a 4 second start time and 0.1 second reading time.
2. Pipet 20 µl of lysed cells per well into a 96-well white (opaque) or black plate, or a luminometer tube.
3. Prime the injector with the QUANTI-Luc™ 4 Reagent solution and proceed with the measurement.

To obtain **end-point readings** using a luminometer **without injectors**.

1. Set the luminometer with a 0.1 second reading time.
2. Pipet 20 µl of lysed cells per well into a 96-well white (opaque) or black plate, or a luminometer tube.
3. Add 50 µl of QUANTI-Luc™ 4 Reagent solution to each well or tube.
4. Gently tap the plate several times to mix (do **not** vortex).
5. Proceed with the measurement.

RELATED PRODUCTS

| Product | Cat. Code |
|--|------------|
| pNiFty2-ELAM-Rluc (NF-κB reporter plasmid) | pnf2-rluc |
| pNiFty3-1-Rluc (IRF reporter plasmid) | pnf3-rluc4 |

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