

QUANTI-Luc™ 4 Lucia/Gaussia

A coelenterazine-based luminescence assay reagent

Catalog code: rep-qlc4lg1, rep-qlc4lg2, rep-qlc4lg5

<https://www.invivogen.com/quantiluc>

For research use only

Version 23G03-AK

PRODUCT INFORMATION

Contents

QUANTI-Luc™ 4 Lucia/Gaussia is a two-component reporter kit which contains:

- QUANTI-Luc™ 4 Reagent (20X)
- QUANTI-Luc™ 4 Stabilizer (25X)

QUANTI-Luc™ 4 Lucia/Gaussia is supplied in different formats:

- rep-qlc4lg1: 1 tube of QUANTI-Luc™ 4 Reagent and 1 tube of QUANTI-Luc™ 4 Stabilizer
- rep-qlc4lg2: 2 tubes of QUANTI-Luc™ 4 Reagent and 2 tubes of QUANTI-Luc™ 4 Stabilizer
- rep-qlc4lg5: 5 tubes of QUANTI-Luc™ 4 Reagent and 5 tubes of QUANTI-Luc™ 4 Stabilizer
- rep-qlc4lg20: 1 bottle of QUANTI-Luc™ 4 Reagent and 1 bottle of QUANTI-Luc™ 4 Stabilizer

Each tube of QUANTI-Luc™ 4 Reagent and QUANTI-Luc™ 4 Stabilizer is sufficient for 5 x 96-well plates (25 ml standard Flash or Glow detection) or 2 x 1536-well plates (8 ml for high throughput screening (HTS)).

Note: For HTS, we recommend to use the Glow detection (see section B in Methods on the next page).

Required Material (not provided)

- Sterile water
- Sterile screw cap tube

Storage and Stability

- Store QUANTI-Luc™ 4 Reagent and QUANTI-Luc™ 4 Stabilizer at -20°C for up to 12 months.
- After preparation, the working solution is stable for 48 hours 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 recombinant Lucia protein or reporter cells.

QUANTI-Luc™ 4 Lucia/Gaussia is a liquid formulation which is highly stable and more convenient to use than the powder formulations QUANTI-Luc™ and QUANTI-Luc™ Gold.

DESCRIPTION

QUANTI-Luc™ 4 Lucia/Gaussia is an optimized kit for the detection of secreted **Lucia** or **Gaussia** activity in live-cell supernatants. This two-component kit comprises QUANTI-Luc™ 4 Reagent (coelenterazine-containing reagent) and QUANTI-Luc™ 4 Stabilizer. 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).

QUANTI-Luc™ 4 Lucia/Gaussia kit provides the user with two options for luciferase activity measurement:

- **Flash (or endpoint) detection:** QUANTI-Luc™ 4 Reagent without QUANTI-Luc™ 4 Stabilizer. This option is ideal for monitoring the cellular response to weak agonists, small sample numbers, or when using microplate readers with reagent injectors.
- **Glow detection:** QUANTI-Luc™ 4 Reagent is used together with QUANTI-Luc™ 4 Stabilizer. This option allows enhanced light signal stability (over 10 min) and is ideal for high-throughput screening (HTS) (i.e. the typical duration to read a 1536-well plate is ~7 minutes) or when using a luminometer without injectors.

PREPARATION OF WORKING SOLUTION

Below are the instructions to prepare QUANTI-Luc™ Lucia/Gaussia working solution for 5 x 96-well plates using the **standard procedure**. Alternatively, you may prepare the working solution from the 20X QUANTI-Luc™ 4 Reagent (and 25X QUANTI-Luc™ 4 Stabilizer) to reach the required volume (see summary table on next page).

Preparation of QUANTI-Luc™ 4 Lucia/Gaussia for **Flash** detection using the **standard procedure**

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 **very briefly** (a few seconds).
3. Use the working solution immediately or store until required for use. QUANTI-Luc™ Lucia/Gaussia "Flash" solution can be stored for 48 hours at 4°C or 1 month at -20°C.

Preparation of QUANTI-Luc™ 4 Lucia/Gaussia for **Glow** detection using the **standard procedure**

1. Dilute the total volume of the 20X tube (1.25 ml) of Reagent and 25X tube (1 ml) of Stabilizer into 22.75 ml sterile water to obtain 25 ml of working solution.
2. Vortex **very briefly** (a few seconds).
3. Incubate at room temperature for at least 15 min.
4. Use the working solution immediately or store until required for use. QUANTI-Luc™ Lucia/Gaussia "Glow" solution can be stored for 48 hours at 4°C or 1 month at -20°C.

TECHNICAL SUPPORT

InvivoGen USA (Toll-Free): 888-457-5873

InvivoGen USA (International): +1 (858) 457-5873

InvivoGen Europe: +33 (0) 5-62-71-69-39

InvivoGen Asia: +852 3622-3480

E-mail: info@invivogen.com

Volumes of Reagent and Stabilizer depending on desired volume of QUANTI-Luc™ 4 Lucia/Gaussia working solution for the standard procedure.

Final volume of working solution	QUANTI-Luc™ 4 Reagent (20X)	QUANTI-Luc™ 4 Stabilizer (25X)	Sterile H ₂ O
For Flash detection (standard procedure)			
5 ml	0.25 ml	-	4.75 ml
10 ml	0.50 ml	-	9.50 ml
25 ml	1.25 ml	-	23.75 ml
For Glow detection (standard procedure)			
5 ml	0.25 ml	0.20 ml	4.55 ml
10 ml	0.50 ml	0.40 ml	9.10 ml
25 ml	1.25 ml	1 ml	22.75 ml

Below are the instructions to prepare QUANTI-Luc™ Lucia/Gaussia working solution for 2 x 1536-well plates using the **high throughput screening (HTS) procedure**.

Preparation of QUANTI-Luc™ 4 Lucia/Gaussia for **Glow** detection using the **HTS** procedure

1. Dilute the total volume of the tubes of Reagent (1.25 ml) and Stabilizer (1 ml) into 5.75 ml sterile water to obtain 8 ml of working solution.
2. Vortex **very briefly** (a few seconds).
3. Incubate at room temperature for at least 15 min.
4. Use the working solution immediately or store until required for use. QUANTI-Luc™ Lucia/Gaussia "Glow" solution can be stored for 48 hours at 4°C or 1 month at -20°C.

METHODS

A. Standard procedure

Flash Detection of luciferase activity from cell culture medium

Prepare the QUANTI-Luc™ 4 Lucia/Gaussia "Flash" working solution as per instructions (see section "preparation of working solution").

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 sample per well into a 96-well white (opaque) or black plate, or a luminometer tube.
3. Prime the injector with the QUANTI-Luc™ 4 Lucia/Gaussia "Flash" solution and proceed **immediately** 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 sample per well into a 96-well white (opaque) or black plate, or a luminometer tube.
3. Add 50 µl of QUANTI-Luc™ 4 Lucia/Gaussia "Flash" solution to each well or tube.
4. Gently tap the plate several times to mix (do **not** vortex).
5. Proceed **immediately** with the measurement.

Glow Detection of luciferase activity from cell culture medium

Prepare the QUANTI-Luc™ 4 Lucia/Gaussia "Glow" working solution as per instructions (see section "preparation of working solution").

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 sample per well into a 96-well white (opaque) or black plate, or a luminometer tube.
3. Prime the injector with the QUANTI-Luc™ 4 Lucia/Gaussia "Glow" solution and proceed **immediately** 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 sample per well into a 96-well white (opaque) or black plate, or a luminometer tube.
3. Add 50 µl of QUANTI-Luc™ 4 Lucia/Gaussia "Glow" solution to each well or tube.
4. Gently tap the plate several times to mix (do **not** vortex).
5. Proceed **immediately** with the measurement.

B. High Throughput Screening (HTS) procedure

The reading of a HTS plate requires enhanced light signal stability. Therefore, we recommend to use the QUANTI-Luc™ 4 Lucia/Gaussia "Glow" solution for this assay (see section "preparation of working solution").

The protocol below has been optimized for HTS screening using 1536 well plates. The QUANTI-Luc™ 4 Lucia/Gaussia working solution is added directly to the cell suspension in HTS plates to reduce liquid handling.

1. Dispense cell suspension and test compounds into a 1536-well white or black plate in a volume that does not exceed 5 µl per well. Incubate cells with test compounds for the desired period of time.
2. Prepare the QUANTI-Luc™ 4 Lucia/Gaussia "Glow" working solution for the HTS procedure as per instructions.
3. Dispense 2 µl of QUANTI-Luc™ 4 Lucia/Gaussia "Glow" solution per well containing ≤5 µl of cell culture in a 1536-well plate.
4. Measure luminescence once the signal has stabilized according to your instrument settings.

RELATED PRODUCTS

Product	Cat. Code
QUANTI-Luc™ 4 Renilla Kit	rep-qlc4r1
pNiFty2-ELAM-Lucia (NF-κB reporter plasmid)	pnf2-lc
pNiFty3-I-Lucia (IRF reporter plasmid)	pnf3-lc
pSELECT-zeo-Lucia™ (expression plasmid)	psetz-lucia
Recombinant Lucia luciferase protein	rec-lucia-1
Reporter Cells	
THP1-Dual™ (IRF-Lucia/NF-κB-SEAP) Cells	thpd-nfis
THP1-Lucia™ NF-κB Cells	thpl-nfkb
Jurkat-Dual™ (IRF-Lucia/NF-κB-SEAP) Cells	jktd-isnf
Jurkat-Lucia™ NFAT Cells	jklt-nfat

TECHNICAL SUPPORT

InvivoGen USA (Toll-Free): 888-457-5873
 InvivoGen USA (International): +1 (858) 457-5873
 InvivoGen Europe: +33 (0) 5-62-71-69-39
 InvivoGen Asia: +852 3622-3480
 E-mail: info@invivogen.com