

Gaussia GLOW-Juice SMALL KIT (1 02 544)

Components include:

Gaussia GLOW-Juice	5x 10 ml Buffer for measuring Gaussia-Luciferase – without substrate. Store at +4°C.
Coelenterazine(CTZ)	5x 1 vial lyophilisated substrate for 10 ml Gaussia GLOW-Juice (synth.) Coelenterazine for Renilla- and Gaussia- Luciferase Store at -80°C in the dark.
Reconstruction buffer	200 µl for dissolving the lyophilisated Coelenterazine Store at -80°C in the dark.
2x Lysis-Juice 2	1 x 10ml dual concentrated Lysis-Buffer without detergents, for measurement of marine luciferases (Renilla/Gaussia) in mammalian cells. Store at +4°C.

Reconstruction:

This Gaussia GLOW-Juice SMALL-KIT includes 5x 10 ml Test-Systems for Gaussia-Luciferase. Note: If the lyophilisated Coelenterazine is dissolved in the Reconstruction buffer the stability will decrease after 30 Days. Please pipette the 200 µl Reconstruction buffer into the brown glass tube of lyophilisated CTZ. It result a **50 x stock solution** that will be stable for at least **30 days** after reconstruction (**store at -80°C!!!**). The calculated amount of Coelenterazine stock solution has to be mixed into the measuring Gaussia GLOW-Juice **shortly before use** (2µl CTZ into 100 µl Gaussia GLOW-Juice).

The reagents should reach at least room temperature (20-25°C) before starting measuring the luciferases!
Remainders of the mixed Gaussia GLOW-Juice should not be frozen again because it will loose noticeable its activity.

Preparation of Cell Lysates:

Gaussia GLOW-Juice SMALL-KIT includes Lysis-Juice 2. This buffer doesn't contain detergence, is dual concentrated and suitable for mammalian cells which were transfected with **Renilla/Gaussia/Firefly Luciferase**. Please dilute the dual concentrated lysis buffer with water or within your cell culture!

Standard protocol for Cells cultured in multiwell-plates

Required final volume of Lysis-Juice 2 per well:

Culture Plate	Vol. Lysis-Juice
6-well	500µl
12-well	250µl
24-well	100µl
48-well	65µl
96-well	20µl

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Standard protocol for Cells cultured in multiwell-plates

- Remove the growth medium from your adherent cells and wash the monolayer two or three times with phosphate buffered saline (PBS)
- Add the required volume of Lysis-Juice to each well (see Tabel)
- Place the plate on a shaker for 15 minutes at room temperature, additional up and down pipetting steps will increase the cell lysis. Freeze cells at -20 or -80°C and thaw them afterwards
- The cell lysate can be placed in storage tubes or measured in the plate by adding reconstructed Gaussia GLOW-Juice.

Standard Protocol

Program luminometer:

For the measurement we suggest a delay of 2 sec. after adding the reagent to the lysate and a measuring time of 5 sec..

- 1.) Transfer 20µl cell lysate into your luminometer tube.
- 2.) Add 100 µl **Gaussia GLOW-Juice**
- 3.) Start measurement

Standard procedure:

