

Renilla-Juice Luciferase Assay

Package Sizes:

Art.no.	Volume	No. Assays
102531	100 ml	(1.000-2.000 assays)
102530	5 x 10 ml	(500-1.000 assays)
102531-1	1 x 10 ml	(100-200 assays)
102531-0	1 x 4 ml	Sample

Components included:

Renilla-Juice	Reaction buffer Store at +4°C.
Coelenterazine	Vial A Store at -20°C.
Reconstruction Buffer	Vial B Store at -20°C.
2x Lysis-Juice	dual concentrated Lysis-Buffer Store at +4°C.

Reconstruction:

Please dissolve the substrate in Vial A with the Reconstruction buffer in Vial B and mix gently. The prepared mixture can be stored at -80 °C. After 30 days the activity of the dissolved substrate will start linear to decrease. Just before measuring the Renilla Luciferase please dilute the substrate mixture in the Reaction Buffer 1:50 (2µl Substrate in 100 µl reaction buffer). Before measuring the buffer should be tempered 37 °C - at least room temperature. Reminders of the mixed Renilla-Juice should not be frozen again because they will loose noticeable activity.

Preparation of Cell Lysates:

Renilla-Juice Luciferase Assay includes a Lysis-Juice. Lysis-Juice is dual concentrated and suitable for mammalian cells which were transfected with Renilla Luciferase. Please dilute the dual concentrated lysis buffer with water or within your cell culture.

Standard Protocol for Cells Cultured in Multiwell Plates

Required volumes of Lysis-Juice

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
- Wash the monolayer two or three times with phosphate buffered saline (PBS)
- Add the required volume of Lysis-Juice to each well (see Table.1)
- Place the plate on a shaker for 15 minutes at room temperature, additional steps of up-down pipetting of cell-buffer mix will increase the lysis (or freeze and defrosting steps could be added)
- Ready to use cell-lysate can placed in storage tubes or measured in the plate by adding Renilla-Juice.

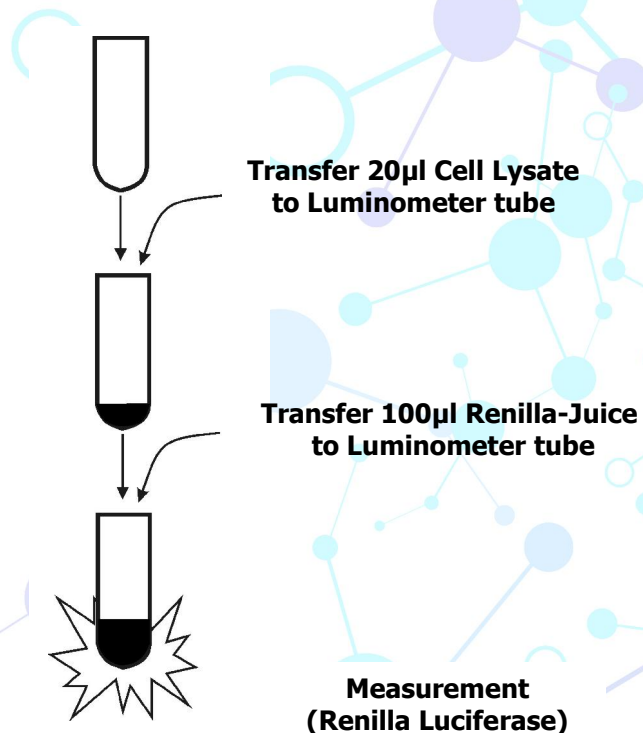
Standard Protocol:

Program Luminometer suggestion: 2 sec. Delay, 5 sec. measurement time (can be customized)

- 1.) Transfer 20 μ l cell lysate to a luminometer tube / microplate well.
- 2.) Add the prepared **Renilla-Juice**.
- 3.) Measurement starts after 2 sec. delay for 5 sec. duration.

Standard procedure:

Program Luminometer
(delay 2sec./measurement duration 5sec)



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