Nov 24, 2022

Luciferase Activity Assay

DOI
https://dx.doi.org/10.17504/protocols.io.dm6gpj37dgzp/v1

Electra Brunialti[1], Alessandro Maria Villa[1], Paolo Ciana[1]

[1]Department of Health Sciences, University of Milan, Milan, Italy

Electra Brunialti

DOI: https://dx.doi.org/10.17504/protocols.io.dm6gpj37dgzp/v1

Protocol Citation: Electra Brunialti, Alessandro Maria Villa, Paolo Ciana 2022. Luciferase Activity Assay. **protocols.io** https://dx.doi.org/10.17504/protocols.io.dm6gpj37dgzp/v1

Protocol status: Working

Created: November 22, 2022

Last Modified: November 24, 2022

Protocol Integer ID: 73113

Keywords: luciferase activity assay luciferase activity, cell extract, using verita, luminometer, assay

Funders Acknowledgements:
The work was supported by the EU Joint Programme - Neurodegenerative Disease Research (JPND) project GBA-PaCTS, 01ED2005B

Abstract

Luciferase activity assay on cell extracts using Veritas (Turner) luminometer.

1 Lysate the cell using 1X Luciferase Cell Culture LysisReagent (Cat. E1531, Promega) following manufacturer instruction;

2 prepare Reaction Buffer (100µl for each sample): 500 µM luciferin (Cat E160E, Promega), 20 mM Tricine, 0.1 mM EDTA, 1.07 mM (MgCO3)4·Mg(OH)2, 2.67 mM MgSO4 in H2O, pH 7.8, with 33.3 mM DTT and 530 mM ATP.

3 put 20 µl/well of cell lysate into the white plate (Cat 3912, Corning);

4 turn on the luminometer (Veritas, Turner);

5 let the samples' plate and Reaction Buffer equilibrate at room temperature; Insert the plate into the instrument and prime the tube with the Reaction Buffer;

6 run the protocol on the luminometer (injection volume 100µl, delay before measurement 0s, integration time 10s)