Cell Meter Glow™ 3D Cell Viability Assay

Catalog number: 21604, 21605, 21606
Unit size: 100 Tests, 1000 Tests, 10000 Tests

Component	Storage	Amount (Cat No. 21604)	Amount (Cat No. 21605)	Amount (Cat No. 21606)
Cell Meter Glow™ 3D Reagent	Freeze (< -15 °C), Minimize light exposure	10 mL (100 Tests)	100 mL (1000 Tests)	10 x 100 mL (10000 Tests)

OVERVIEW

Cell Meter Glow™ 3D Cell Viability Assay is designed for measuring viable cells in three-dimensional culture systems using a homogeneous luminescent ATP readout. ATP levels reflect the presence of metabolically active cells, and luciferase-based detection converts ATP-dependent activity into a luminescent signal that can be measured with a standard luminescence microplate reader. The assay format is intended for 3D culture models, including spheroids, organoids, and cells grown in extracellular matrix-supported environments. Its reagent system is formulated to support cell disruption and ATP recovery from dense 3D structures, enabling endpoint viability analysis in microplate workflows. Cell Meter Glow™ 3D Cell Viability Assay is suitable for 3D cell viability analysis, compound response studies, cytotoxicity testing, and high-throughput screening applications.

AT A GLANCE

Protocol Summary

1. Prepare 3D cell culture samples with test compounds
2. Add equal volume of ready-to-use Cell Meter Glow™ 3D Reagent (100 µL/96-well plate or 25 µL/384-well plate)
3. Mix thoroughly to allow reagent contact with the 3D culture sample
4. Incubate at room temperature for 10 - 30 minutes
5. Monitor the luminescence intensity

Important Note

To achieve the best results, it's strongly recommended to use the white plates. Thaw all the kits components at room temperature before starting the experiment.

KEY PARAMETERS

Luminescence microplate reader

Excitation	--
Emission	--
Instrument specification(s)	Solid white

CELL PREPARATION

Prepare 3D cell culture samples according to the specific experimental model. Samples may include spheroids, organoids or cells cultured in compatible extracellular matrix-supported formats.

For compound response or cytotoxicity studies, treat samples with test compounds for the desired period of time before adding Cell Meter Glow™ 3D Cell Viability Reagent.

Note: Assay conditions may need to be optimized depending on cell type, 3D culture size, plate format, matrix composition and treatment conditions.

PREPARATION OF STANDARD SOLUTIONS

For convenience, use the Serial Dilution Planner:
https://www.aatbio.com/tools/serial-dilution/21604

ATP standard
This is optional, ATP is not provided. Make 1 mM ATP stock solution with ddH2O or appropriate buffer, then perform serial dilution to achieve ATP concentrations ranging from 10 pM to 10 µM.

SAMPLE EXPERIMENTAL PROTOCOL

Table 1. Layout of ATP standards and test samples in a white 96-well microplate. SD=ATP Standard, BL=Blank Control, TS=Test Sample

BL	BL	TS	TS
SD1	SD1	...	...
SD2	SD2	...	...
SD3	SD3		
SD4	SD4		
SD5	SD5		
SD6	SD6		
SD7	SD7		

Table 2. Reagent composition for each well

Well	Volume	Reagent
SD1-SD7	100 µL	Serial Dilution of ATP (e.g. 10 pM to 10 µM)
BL	100 µL	Cell Meter Glow™ 3D Reagent
TS	100 µL	3D Cell Culture Sample

1. Prepare 3D cell culture samples in a white 96-well plate or 384-well plate.
2. Treat cells (or samples) with test compounds by adding 10 µL of 10X compounds for a 96-well plate or 5 µL of 5X compounds for a 384-well plate in desired compound buffer. For blank wells (medium without the cells), add the corresponding amount of compound buffer.
3. Incubate the cell plate in a 37°C, 5% CO2 incubator for the desired period of time, such as 24, 48 or 96 hours.
4. Equilibrate the cell plate and Cell Meter Glow™ 3D Reagent to room temperature before use.
5. Add an equal volume of Cell Meter Glow™ 3D Reagent to each well, mix well to allow reagent contact with the 3D sample.
   - For a 96-well plate, add 100 µL/well of Cell Meter Glow™ 3D Reagent to 100 µL/well of sample.
   - For a 384-well plate, add 25 µL/well of Cell Meter Glow™ 3D Reagent to 25 µL/well of sample.
6. Incubate at room temperature for 10 - 30 minutes.
7. Monitor the luminescence intensity with a standard luminometer.

Note: Aliquot and store the unused Cell Meter Glow™ 3D Cell Viability Reagent at -20 °C, and avoid repeated freeze/thaw cycles.

EXAMPLE DATA ANALYSIS AND FIGURES

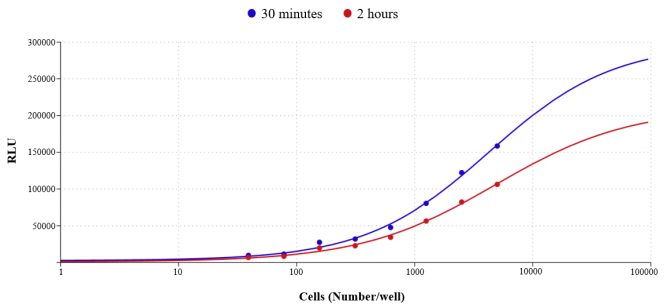

Figure 1. Luminescent response of 3D-cultured HeLa cells measured using the Cell Meter Glow™ 3D Viability Assay Kit. HeLa cells were plated as a 2-fold serial dilution in Greiner LUMITRAC™ 200 96-well microplates and luminescence was measured using a CLARIOstar plate reader (BMG LABTECH) after 15 minutes and 2 hours of incubation. The assay detected as few as 39 cells under the tested conditions. Luminescence increased with cell number and the response curves were fitted using a four-parameter logistic (4PL) model. Signal was retained for at least 2 hours under the assay conditions.

DISCLAIMER

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