PROTOCOL

Evaluation of cell viability

— Using CellTiter-Glo 3D cell viability assay

Application

This protocol explains how to evaluate cell viability using CellTiter-Glo 3D cell viability assay with P3D Scaffolds. For more protocols, please visit our Resources Platform.

This assay lyses all cells and measures the amount of ATP present. It works by mixing cells in media 1:1 with Assay Reagent and measuring output luminescence. The output requires the use of black/opaque 96-well plates with clear bottoms. This procedure describes how to use this kit with scaffolds. Please also refer to manufacturer Promega's protocol.

Plate type	Media volume	Assay Reagent volume
96 well plate	100 uL	100 uL
24 well plate	200 uL	200 uL
12-well plate	500uL	500uL
6-well plate	1500uL	1500uL

Always include cell-free scaffolds as a negative control.

Materials

- CellTiter-Glo 3D cell viability assay
- Plate shaker
- Opaque plate
- Plate reader

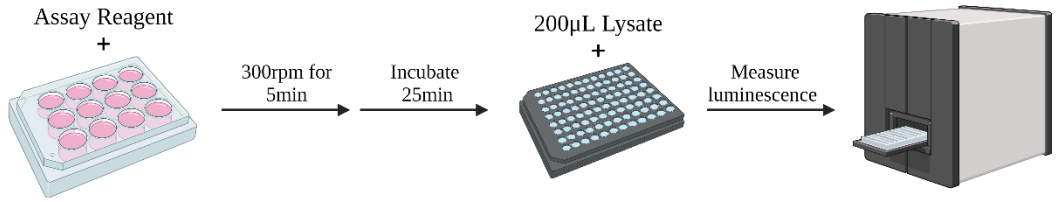

FIGURE 1: *WORKFLOW FOR EVALUATING CELL VIABILITY USING CELLTITER-GLO 3D CELL VIABILITY ASSAY.*

Notes before starting and general advice on material handling

- All handling of The P3D Scaffolds products should be performed using gloves, according to the standard aseptic methods.
- The scaffolds are supplied sterile by dry heat sterilization and remains sterile until opened.

Procedure

The day before performing the procedure: place Assay Reagent at 4C to allow for slow thawing

Before you start

- Place Assay Reagent and cell plates at RT to equilibrate for at least 30 min
- Gently invert assay reagent bottle to mix before use
- Make aliquot of assay reagent by calculating the amount needed
- If necessary, remove media from wells to achieve volumes listed in table above

1. Add Assay Reagent to each well, using multichannel pipette if possible. Volume of Assay Reagent should be equal to the volume media already present in wells

2. Place plate at RT on a plate shaker at 300 rpms for 5 min

3. Remove plate from plate shaker and incubate plate at RT for 25 min

4. Pipette 200 uL lysate to opaque plate as quickly as possible

5. Measure luminescence using a plate reader with integration time 0,25-1 seconds. Settings may vary between instruments

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Patent status:

The product is protected by one or more US, European, and/or foreign patents.

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