

## ab284576 – Cell Proliferation Assay Kit (Fluorometric)

For the measurement of cell proliferation and cytotoxicity.  
For research use only - not intended for diagnostic use.

For overview, typical data and additional information please visit:

<http://www.abcam.com/ab284576>

### Storage and Stability

On receipt entire assay kit should be stored at -20°C, protected from light. Kit has a storage time of 1 year from receipt, providing components have not been reconstituted.

### Materials Supplied

Item	Quantity	Storage Condition
Cell Lysis (5X)	25 mL	-20°C
Nuclear Dye (200X)	0.65 ml	-20°C

### Materials Required, Not Supplied

- 96-well white tissue culture plate with clear bottom
- Multichannel or single channel Pipettes
- Multi-well spectrophotometer (Fluorescence reader)

### Reagent Preparation

- Before using the kit, spin the tubes prior to opening.

Cell Lysis Buffer (5X): Store at 4°C or -20°C. Bring to room temperature before use

Nuclear Dye (200X): For long-term storage, aliquot, and store at -20°C. Avoid freeze/thaw.

### Assay Protocol

#### Sample Preparation

1. Culture cells (0.1 – 5 x 10<sup>4</sup> cells/well) in a 96-well white tissue culture plate in a final volume of 100 µL/well medium in the absence or presence of testing compounds. Incubate cells for the desired time.

#### Δ Notes:

- a. For adherent cells, seed the cells one day before treating with testing compounds.
  - b. Incubation time depends on the testing compounds.
  - c. For toxicity assay, use more cells (e.g., 5 – 8x10<sup>4</sup> cells/well).
  - d. (Optional) For low cell number and/or higher sensitivity, after incubation period, centrifuge cells at 400 x g for 5 min and carefully remove supernatant using multichannel pipette (Don't use vacuum aspirator). Dilute Nuclear Dye (200X) with Cell Lysis Buffer (5X) and dH<sub>2</sub>O to make 1X Nuclear Dye/Cell Lysis Buffer solution. Add 100 µL of 1X Nuclear Dye/Cell Lysis Buffer solution into each well. Gently shake the plate for 15 min. on a shaker at room temperature protected from light and directly proceed to step 3.
2. Prepare 25 µL/well of 5X Nuclear Dye/Cell Lysis Buffer solution by adding 0.625 µL of Nuclear Dye (200X) to 25 µL of Cell Lysis Buffer (5X). Make as much as needed. Mix. After incubation, directly add into each well. Gently shake the plate for 15 min on a shaker at room temperature protected from light.
  3. Measure fluorescence of treated and untreated cells using a microtiter plate reader at Ex/Em = 480/538 nm.

#### Δ Notes:

- a. Assay has a linear range up to 60,000 cells. Depending on the cell type, adjust the cell number to make sure that fluorescence reading is within the linear range.
- b. Serial dilutions of testing cells can be used to make standard curve.

### **Technical Support**

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