



## MTT Cell Viability & Proliferation Assay

### Cat. No. CB016

(1000 Tests in 96-well plate)

### Product Description

The study of cell viability and proliferation is very important for evaluating a cell population's responses to external factors, such as growth factors, antibiotics and anti-cancer drugs. The MTT Cell Viability & Proliferation Assay allows simple, accurate and reliable counting of metabolically active cells, based on the conversion of pale yellow tetrazolium MTT [3-(4,5-Dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide] to purple formazan crystals. The crystals can be solubilized and then spectrophotometrically quantified.

### Kit Components

MTT, 5 vials, 10 mg, 5 mg/mL

MTT Solubilization Buffer, 100ml, 0.04-0.1 N HCl in isopropanol

Typical use: Add 1/10th of culture volume

### Procedures (96-well plate)

1. Plate and culture cells in a clear-bottom 96-well tissue culture plate. Incubate cells with test compounds and controls for the desired period of time. The final volume of culture medium in each well should be 100  $\mu$ l.
2. Reconstitute each vial of MTT with 2 ml of PBS, pH 7.4. Vortex briefly, sterile filter and keep in the dark at 4°C until use. Fresh reconstitution of MTT is recommended for each experiment, although reconstituted MTT solution should be stable for up to 2 weeks when kept at 4°C, protected from light. (Filter reconstituted MTT solution to remove residues/precipitates)
3. Equilibrate the MTT Solution to room temperature, and then add 10  $\mu$ l of MTT Solution to each well (the volume of MTT solution should be 1/10 of the original culture medium). Mix well by gently rocking the plate side-to-side.
4. Incubate cultures with MTT at 37°C for 2-4 hours depending on cell type and seeding density. At the end of incubation, there should be black crystals formed in the live cells.
5. After incubation, add 100  $\mu$ l of MTT Solubilization Buffer (equal to the volume of original culture medium) to each well and pipette up and down to help dissolve crystals. Gentle mixing on an orbital shaker will further enhance dissolution.
6. Within an hour, measure the absorbance on an ELISA plate reader with a test wavelength at 570 nm and a reference wavelength at 690 nm, and subtract the 690 nm background absorbance from the 570 nm measurement.

### Usage

MTT Cell Viability & Proliferation Assay is used to coat cell culture vessels *in vitro*. MTT Cell Viability & Proliferation Assay is for research use only. It is not approved for human or animal use, or application in clinical or *in vitro* diagnostic procedures.