

# MTT Cell Proliferation and Cytotoxicity Assay Kit

Cat: M1020

Size: 500T/1000T

Storage: MTT Solution should be stored at -20°C; Formazan Dissolving Solution can be stored

at room temperature or -20°C condition. The period of validity is one year.

### **Product content:**

Component	500T	1000T
MTT Solution	5mL	5mL×2
Formazan Dissolving Solution	60mL	60mL×2

## **Introduction:**

The kit is designed for the spectrophotometric measurement of cell growth as a function of mitochondrial activity in living cells. Using MTT could measure the activity of living cells via mitochondrial dehydrogenase activity with simple, accurate and reproducible functions. Dehydrogenases in mitochondria can deoxidize MTT to crystalline purple product Formazan. The crystals can be dissolved in specific solvent. Use Microplate Absorbance Reader to measure absorbance near 490nm wavelength. An increase in cell number results in an increase in the amount of MTT Formazan formed and an increase in absorbance.

For best results, cell numbers should be determined during log growth stage. Each test should include a blank containing complete culture medium without cells.

#### **Protocols:**

- 1. Harvest cell(log growth stage) adjust the concentration of cell suspension, separate into 96 well plate,  $180\mu\text{L/well}$ , 3000-10000 cell/well.
- 2. Return cultures to incubator (37°C, 5%CO2) and incubate for 6 to 24 hours.
- 3. Add appropriate concentrations of toxicant into the culture media in the plate. Incubate the plate for an appropriate length of time in the incubator.
- 4. Remove supernatant, add  $90\mu L$  fresh culture medium then add  $10\mu L$  MTT Solution then continue to culture for 4h.
- 5. Remove supernatant, each well added with  $110\mu L$  Formazan Dissolving Solution, Slowly stirring in a gyratory shaker for 10min, make the crystal is fully dissolved. Measure the absorbance at 490nm using a microplate reader.
- 6. Meanwhile set the zero adjustment holes (Culture medium, MTT Solution, Formazan Dissolving Solution), control holes (Cell, solvent of the same concentration toxican, Culture medium, MTT Solution, Formazan Dissolving Solution), each group set 3 holes.

#### Notes:

- 1. Uneven evaporation of culture fluid in 96well plates may cause erroneous results. The outermost circle is most easy to evaporate, so you can discard them.
- 2. MTT Solution is stable when stored frozen. Storage at 2-8°C may result in decompositionand yield erroneous results. MTT Solution should be storage avoid light, as MTT will invalidation when



light for a long time.

3. Development of dark color or formation of crystals indicate product deterioration. If the Formazan Dissolving Solution is frozen or precipitate can be dissolved in 37°C water bath, the precipitate must be dissolved and mixed before use.

# **Related products:**

12100	DMEM(H)
31800	RPMI Medium 1640
24800	Cryopreservation Medium
P1020	1×PBS (PH7.2-7.4, 0.01M)
H1020	Hanks(with calcium, magnesium, phenol red)(HBSS)
T1300	Trypsin-EDTA solution, 0.25% (without phenol red)
H1095	1M Hepes (pH7.2-7.4, sterile)
P1400	Penicillin-Streptomycin Liquid