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ISO 13485:2016 ISO 9001:2015

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MTT Cell Proliferation and Cytotoxicity Assay Kit

M1020 (500Test)

FOR RESEARCH USE ONLY, DO NOT USE IT IN CLINICAL DIAGNOSIS

Product Description

MTT can be reduced by some dehydrogenases in the mitochondria to form a crystalline, dark purple product called formazan. In the presence of a specific solvent, it can be completely dissolved. The absorbance at wavelengths near 490nm can then be measured using a microplate reader. The more and faster the cell proliferation, the higher the absorbance; the greater the cell toxicity, the lower the absorbance.

Kit components

Reagent	Volume	Storage
MTT Solution	5mL	-20°C
Formazan Dissolution Solution	6mL	RT

Storage: Kit can be kept at -20°C, protected from light, and is valid for 1 year

Protocol (for reference only)

1. Collect logarithmic phase cells, adjust the cell suspension concentration, and seed them in a 96-well plate, with 180µL per well, at a density of 3000-10000 cells per well.
2. Incubate at 37°C with 5% CO₂ to allow cell adherence, for 6-24 hours.
3. Add the test compounds at appropriate concentrations and continue incubation for an appropriate period of time.
4. Carefully aspirate the supernatant, add 90µL of fresh culture medium, and then add 10µL of MTT solution. Continue incubation for 4 hours.
5. Then aspirate the supernatant and add 110µL of Formazan dissolution solution to each well. Place on a shaker and agitate at low speed for 10 minutes to fully dissolve the crystals. Measure the absorbance of each well at 490nm using an enzyme-linked immunosorbent assay (ELISA) plate reader.
6. Set up blank wells (medium, MTT, Formazan dissolution solution) and control wells (cells, same concentration of drug solvent, medium, MTT, Formazan dissolution solution), with three replicate wells per group.

Notes:

1. Since a 96-well plate is used for the assay, if the cell culture period is relatively long, it is essential to pay attention to the issue of evaporation. On the one hand, as the outermost ring of wells in a 96-well plate is most susceptible to evaporation, one can choose to leave this ring unused and instead fill it with PBS, water, or culture medium. On the other hand, placing the 96-well plate near the water source inside the incubator can help mitigate evaporation.
2. The MTT solution is yellow and needs to be protected from light, as prolonged exposure can lead to its inactivation. Do not use it if the colour changes to grayish-green. If the Formazan dissolution solution freezes or forms a precipitate, it can be incubated in a 37°C water bath to promote dissolution. It must be used only after it has completely dissolved and been thoroughly mixed.
3. The product information is for reference only.
4. The products are all for scientific research use only. Do not use it for medical, clinical diagnosis or treatment, food and cosmetics, etc. Do not store them in ordinary residential areas
5. For your safety and health, please wear laboratory clothes, disposable gloves and masks to operate.