CPT-0103

MTT Cell Proliferation Assay

(1000 wells, Colorimetric, OD 590 nm, Store at -20°C)

Introduction:

The ability to quantify the number of viable cells in a system under controlled conditions is a requirement in cell biology research. There are several such assays designed to measure metabolic activity of cultured cells, of which MTT is one. The advantages of AkrivisBio's MTT Cell Proliferation Assay are sensitivity (subtle changes can be observed), cost-effectiveness, versatility (adherent or suspension cultures, 96 or 384 well plates) and the ability to monitor metabolic activity over long periods of time. The assay is based on the conversion of MTT to an insoluble formazan. Cells with active metabolism convert MTT while dead cells can't. Absorbance at 590 nm is proportional to metabolic activity. While this is typically due to the number of viable cells, it can be due to increased metabolic activity in the same number of cells. This assay provides an easy-to-use, non-radioactive, and high-throughput method for cell proliferation, cell viability, chemotaxis, cytotoxicity, and apoptosis.

Applications:

Studying the metabolic effects of growth factors, cytokines, mitogens, and nutrients
Studying the cytotoxicity of anticancer drugs and other cytotoxic agents

Assay Components:

MTT Reagent	50 ml	NM	CPT-0103A
MTT Solvent	100 ml	NM	CPT-0103B

User supplied Materials:

Serum-free culture medium

Storage Handling Considerations:

Store unopened assay at -20°C.
MTT Reagent: Ready to use as supplied. MTT is light sensitive. It's best to use it under subdued light conditions. Open under sterile conditions. It can be stored at 4°C for up to 2 months. For longer term storage, keep it at -20°C. Bring to room temperature before use.
MTT Solvent: Ready to use as supplied. Store at room temperature.

Assay Protocol:

1. Grow cells at the required densities for the cell type under study, on a clear plate. Dissolve compounds of interest at high concentration in an appropriate solvent. Apply compound solution to cells, keeping solvent concentration as low as possible for the desired time period. Prepare paired solvent control wells for each test well using the same final solvent concentration. For adherent cells, carefully discard the culture medium. For suspension cells, spin the plate gently at 1,000 X g and 4°C for 5 minutes then carefully discard the culture medium. Keep one empty well available as a reagent control well.
2. Each well will require 100 µl of MTT mix. Prepare sufficient MTT mix for the number of wells (test, solvent control, and reagent control) to be analyzed. For each well, mix 50 µl of serum-free medium and 50 µl of MTT Reagent. To the empty reagent background control, add 100 µl of MTT Reagent Mix. Incubate the plate for 3 hours at 37°C.
3. After incubation, add 150 µl of MTT Solvent to each well. Protect the plate from light by wrapping it in aluminum foil and place on an orbital shaker for 15 minutes, then read absorbance at 590 nm.

FOR RESEARCH USE ONLY! Not to be used for diagnostic or therapeutic purposes.

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