FOR RESEARCH USE ONLY! Not to be used on humans

SuperSensitive™ Cell Proliferation Assay Kit (Colorimetric)

(Catalog # F6004-1000; Store at -20°C)

Material Supplied

Item #	Item	Quantity	Storage
F6004-1000-A	SuperSensitive Reagent	1 Vial	-20°C
F6004-1000-B	Electro Mediator Solution (EMS)	10 ml	-20°C

Introduction

BioIntersect's SuperSensitive™ Cell Proliferation Assay uses WST-8, which is more stable and less toxic to the cells compared to other tetrazolium salts like MTT, WST-1 or XTT. Mitochondrial dehydrogenases in the cells, reduces WST-8 and produces water-soluble formazan that directly dissolves in culture media. Number of viable cells are directly proportional to the mitochondrial dehydrogenases activity, and the amount of formazan dye formed, which is measured by absorbance at 450 nm. BioIntersect's SuperSensitive™ Cell Proliferation Assay is a colorimetric assay for assessing cell viability, cytotoxity and proliferation. Our kit provides a sensitive, easy-to-do, one-step, non-radioactive and high-throughput method for cell proliferation, cell viability, chemotaxis, cytotoxicity, and apoptosis.

Samples

- Proliferating and non-proliferating cultured cells.

Applications

- Measurement of cell viability in response to growth factors, toxic compounds, mitogens, cytokines and nutrients.
- Analysis of anticancer drugs and other pharmaceuticals.
- Assessment of cell cycle and biological mediators that affect cell growth

Pre-Assay Preparations

Store kit at -20°C, protected from light. Stable for over 1 year.
SuperSensitive™ Reagent: Dissolve the reagent into 2 ml of Electro Mediator Solution (EMS). Aliquot as needed (1 ml is sufficient for one 96-well plate assay) and store at –20°C to avoid freeze/thaw. Read the entire protocol before performing the assay. Use clear plates for the assay.

Assay Protocol

1. **Sample Preparation:** Grow cells of interest in a 96-well clear plate in a final volume of 100 µl/well, according to the desired protocol. Cells seeded at densities between 5,000-10,000 cells/well should reach optimal population densities within 48-72 hrs. Grow ~50,000 cells/well for 24 hrs assay. For toxicity studies, use > 50,000 cells per well. Treat the cells with compounds/drugs of interest in an appropriate solvent for desired period of time. We recommend treat parallel well(s) as solvent control and use same volume of solvent as for the treated cells.
2. **Procedure:** Add 10µl of SuperSensitive™ Reagent into each well. For background control, add 10 µl of SuperSensitive™ Reagent into a well containing media only (no cells). Incubate the plate for 3-4 hrs at 37°C. We recommend determining optimal incubation time for a particular experiment depending on the cell type and cell number.
3. **Reading and Calculations:** Before reading, we recommend shaking the plate in an orbital shaker for 1 minute. Read the 96-well plate absorbance at 450 nm as endpoint setting. Subtract the absorbance of the control well from all absorbance value to yield Corrected Absorbance Values. Plot Corrected Absorbance Values 450 nm (Y axis) versus concentration of growth factor/Cytotoxic agent (X axis, log scale). Determine the ED50/IC_{50} value by locating the X-axis value corresponding to one-half the maximum (plateau) absorbance value.

Notes: If comparing the affect of several compound(s) on the cell viability, plot the absorbance (Y-axis) and compare different treatments.

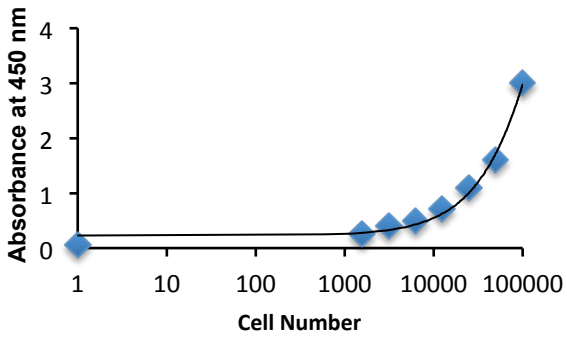

Figure: SuperSensitive™ Cell Proliferation Assay: HeLa cells were serially diluted in a 96-well clear plate and incubated for 4 hr with SuperSensitive™ Reagent at 37°C, as described in the protocol. Absorbance was measured at 450 nm and Abs is plotted as the function of Cell Number.

RELATED PRODUCTS:

MTT Cell Proliferation Assay Kit (Colorimetric)
MTS Cell Proliferation Assay Kit (Colorimetric)
WST-1 Cell Proliferation Assay Kit (Colorimetric)
Calcein AM Cell Proliferation Assay Kit (Fluorometric)
SetuBlue™ Cell Proliferation Assay Kit (Fluorometric)
Cholesterol Efflux Assay (Cell Based Fluorometric)
Glucose Uptake Assay (Cell Based Colorimetric)
Mammalian Protein Extraction Buffer
Bacterial Protein Extraction Buffer
Membrane Protein Extraction Buffer
Yeast Protein Extraction Buffer
Tissue Protein Extraction Buffer

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