
Product Manual

CytoSelect™ WST-1 Cell Proliferation Assay Reagent

Catalog Number

CBA-253

960 assays in 96-well plate format

FOR RESEARCH USE ONLY
Not for use in diagnostic procedures



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Creating Solutions for Life Science Research

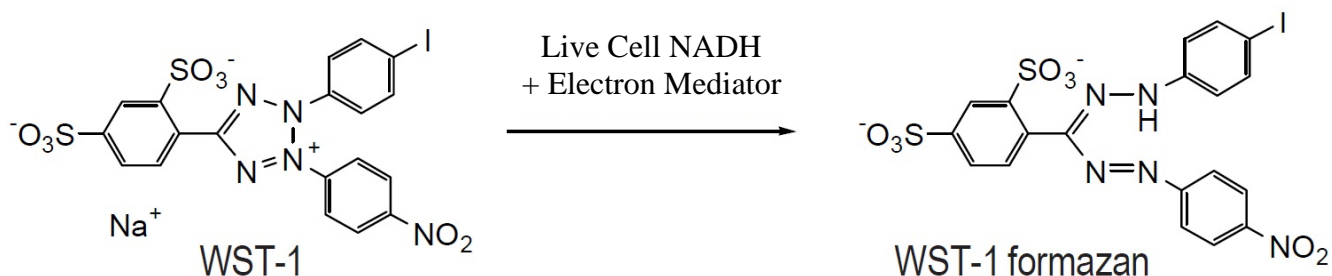
Introduction

The measurement and monitoring of cell proliferation is an essential technique in any laboratory focused on cell-based research. This skill allows for the optimization of cell culture conditions as well as the determination of cytokine, growth factor, or hormone activity. More importantly, the cytostatic nature of anticancer compounds in toxicology testing, the efficacy of therapeutic chemicals in drug screening, and cell-mediated cytotoxicity can all be assessed through the quantification and monitoring of cell proliferation.

Cell proliferation characteristics include cellular metabolic activity and cell membrane integrity. One method for measuring metabolic activity is to incubate the cells with a tetrazolium salt such as MTT, which is cleaved into a colored formazan product by metabolically active cells. Similarly, the green fluorescent dye Calcein AM can measure intracellular esterase activity in proliferating live cells, which is another indicator of cell viability.

Assay Principle

Cell Biolabs' CytoSelect™ WST-1 Cell Proliferation Assay Reagent provides a colorimetric format for measuring and monitoring cell proliferation. The 10 mL volume is sufficient for the evaluation of 960 assays in ten 96-well plates or 192 assays in eight 24-well plates. Cells can be plated and then treated with compounds or agents that affect proliferation. Cells are then detected with the proliferation reagent, which is converted in live cells from WST-1 to the formazan form in the presence of cellular NADH and an electron mediator.



An increase in cell proliferation is accompanied by increased signal, while a decrease in cell proliferation (and signal) can indicate the toxic effects of compounds or suboptimal culture conditions. The assay principles are basic and can be applied to most eukaryotic cell lines, including adherent and non-adherent cells and certain tissues. This cell proliferation reagent can be used to detect proliferation in bacteria, yeast, fungi, protozoa as well as cultured mammalian and piscine cells.

Related Products

1. CBA-080: CytoSelect™ 24-Well Anoikis Assay
2. CBA-081: CytoSelect™ 96-Well Anoikis Assay
3. CBA-230: Cellular Senescence Assay Kit (SA-β-gal Staining)
4. CBA-231: 96-Well Cellular Senescence Assay (SA β-Gal Activity)
5. CBA-232: Quantitative Cellular Senescence Assay (SA β-Gal)
6. CBA-240: Cell Viability and Cytotoxicity Assay
7. CBA-250: CytoSelect™ Fluorometric Cell Proliferation Assay Reagent

Materials Not Supplied

1. Cells for measuring proliferation
2. Cell culture medium
3. 24-well or 96-well clear cell culture plates.
4. Microtiter plate reader capable measuring absorbance at 450 nm.

Storage

CytoSelect™ WST-1 Cell Proliferation Assay Reagent is a clear, slightly red, ready-to-use solution and should be stored at 4°C. If precipitates or turbidity are observed upon thawing, warm the solution to 37°C for 5–10 minutes and agitate to dissolve the precipitates. For longer periods, aliquot and store at –20°C protected from light to avoid repeated freeze-thaw cycles.

Assay Protocol

1. Prepare a cell suspension containing 0.1-1.0 x 10⁶ cells/ml in medium.
2. Add 100 μL of cell suspension per well to a 96-well cell culture plate or 500 μL per well to a 24-well cell culture plate with or without the compound to be tested. Culture the cells for 24-96 hours at 37°C and 5% CO₂ in a humidified incubator.
3. Add 10 μL of the CytoSelect™ WST-1 Cell Proliferation Assay Reagent to each well if using a 96-well plate, or 50 μL to each well of a 24-well plate.
4. Incubate the plate at 37°C and 5% CO₂ for 0.5 to 4 hours.
5. Read absorbance using 450 nm as the primary wave length.

Example of Results

The following figure demonstrates typical results with the CytoSelect™ WST-1 Cell Proliferation Assay Reagent. One should use the data below for reference only. This data should not be used to interpret actual results.

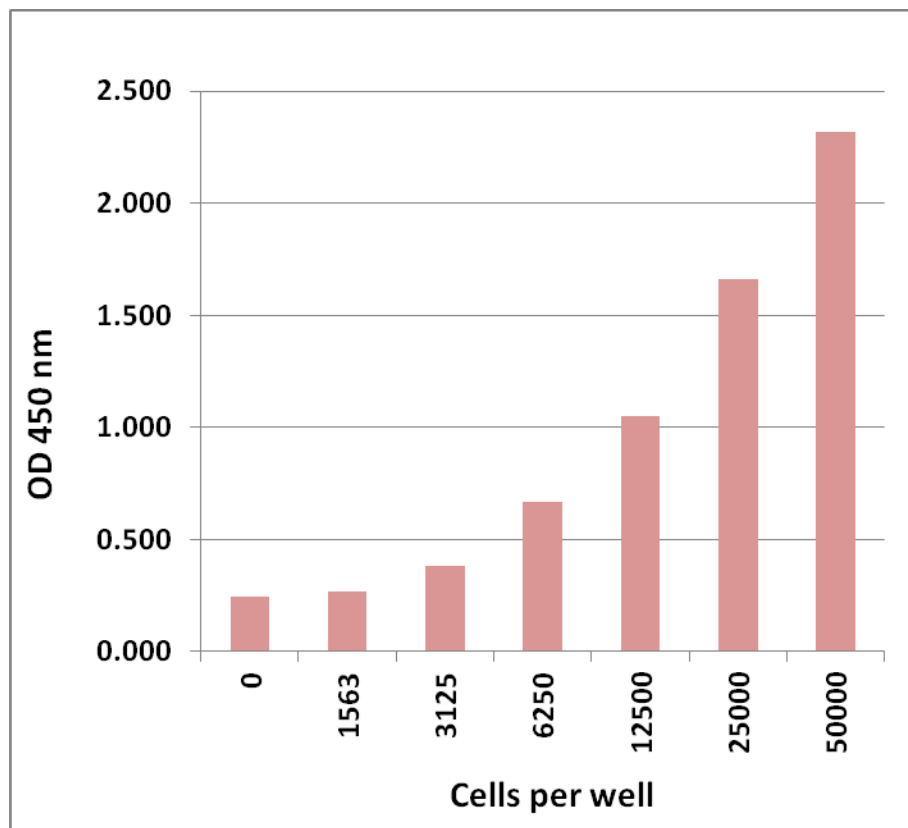


Figure 1. Human HEK 293 Cell Density. HEK 293 cells were seeded at various densities as indicated above and allowed to grow for 24 hours. After adding the CytoSelect™ Cell Proliferation Assay Reagent, cells were then incubated for 2 hours at 37°C and 5% CO₂.

References

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5. Weil M, Jacobson MD, Coles HS, Davies TJ, Gardner RL, Raff KD, Raff MC. (1996) *J Cell Biol* **133**, 1053.
6. Zurgil N, Shafran Y, Fixler D, Deutsch M. (2002) *Biochem Biophys Res Commun* **290**, 1573.

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