Ab228554 – Cell Counting Kit 8 (WST-8)

For the measurement of cell viability of live cells.
This product is for research use only and is not intended for diagnostic use.

For overview, typical data and additional information please visit: www.abcam.com/ab228554 (use www.abcam.cn/ab228554 for China, or www.abcam.co.jp/ab228554for Japan)

Overview

The cell Counting Kit 8 (WST-8) (ab228554) is a convenient and robust way of measuring cell viability. The kit uses a water-soluble tetrazolium salt to quantify the number of live cells by producing an orange formazan dye upon bio-reduction in the presence of an electron carrier.

Materials Supplied:

Item	Quantity	Storage temperature (before prep)
WST-8 Solution	1 bottle (10 mL)	4°C

Storage and Stability:

Store kit at 4°C in the dark immediately on receipt and check below for storage for individual components. Kit can be stored at 4°C for 1 year from receipt, if components have not been reconstituted.

Store at -20°C for longer-term storage.

Avoid repeated freeze-thaws of reagents.

Materials Required, Not Supplied

These materials are not included in the kit, but will be required to successfully perform this assay:

- Microplate reader capable of measuring absorbance at OD = 460 nm
- 96 well plate with clear flat bottom.

Precautions

Please observe safe laboratory practice and consult the safety datasheet.

For general guidelines, precautions, limitations on the use of our assay kits and general assay troubleshooting tips, particularly for first time users, please consult our guide: www.abcam.com/assaykitguidelines

Reagent Preparation:

WST-8 Solution: Ready to use as supplied.

Assay Procedure

Equilibrate all materials and prepared reagents to room temperature just prior to use and gently agitate.

1. **Cell Proliferation and Cytotoxicity Assay**
   - Plate 5000 to 10,000 cells per well in a tissue culture microplate with a clear bottom.
   - Add test compounds into cells and incubate for a desired period (e.g. 24, 48 or 96 hours) in a 37°C, 5% CO2 incubator.
   - For blank wells (medium without cells), add the same amount of test compounds. The suggested volume is 100 µl for a 96 well plate and 50 µl for a 384 well plate.
   - **Note**: Each cell line should be evaluated on an individual basis to determine the optimal cell density for proliferation or cytotoxicity induction. For proliferation assays, use fewer cells; for cytotoxicity assays, use more cells to start with.
   - Add 10 µl/well (96 well plate) or 5 µl/well (384-well plate) of WST-8 Solution to each well. Protect from the light and incubate for 1-4 hours at 37°C.
   - **Note**: The incubation time could be from 30 minutes to overnight depending on the individual cell type and cell concentration used. Optimize the incubation time for each experiment.
   - Measure the absorbance increase at 460 nm.

2. **Cell Counting assay**
   - Prepare cell culture in a tissue culture microplate with clear bottom. The suggested total volume is 100 µL for a 96-well plate, and 50 µL for a 384-well plate.
   - Add 10 µl/well (96 well plate) or 5 µl/well (384-well plate) of WST-8 Solution to each well. Protect from the light and incubate for 1-4 hours at 37°C.
   - **Note**: The incubation time could be from 30 minutes to overnight depending on the individual cell type and cell concentration used. Optimize the incubation time for each experiment.
   - Measure the absorbance increase at 460nm.

Typical Data

Data provided for demonstration purposes only.

- The absorbance of the blank wells with only growth medium is subtracted from the values for those wells with cells.

  - **Note**: The absorbance of the blank wells may vary depending on the sources of the microtiter plates or the growth media.
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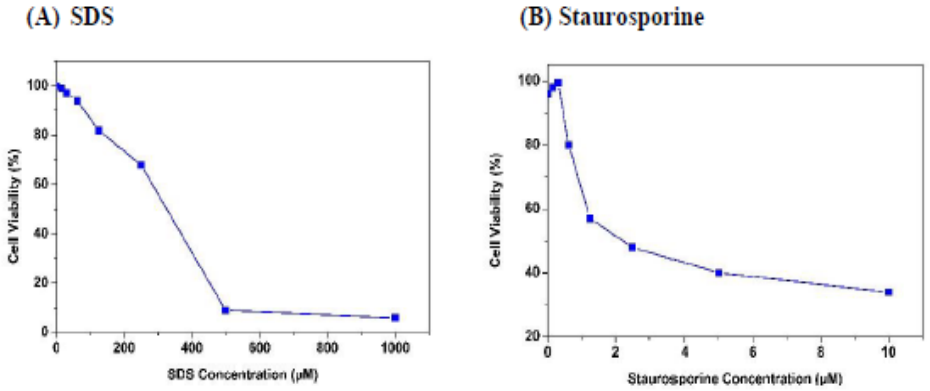

Figure 1: Cytotoxicity tests of HeLa cells in response to (A) SDS and (B) Staurosporine treatment were measured with the cell Counting Kit 8 (WST-8) (ab228554). HeLa cells at 10,000 cells/well/100 µL were seeded overnight in a black wall/clear bottom 96-well plate. Cells were treated with serially diluted SDS for 2 hours or Staurosporine for 4 hours. The absorbance was measured at 460 nm using a plate reader.

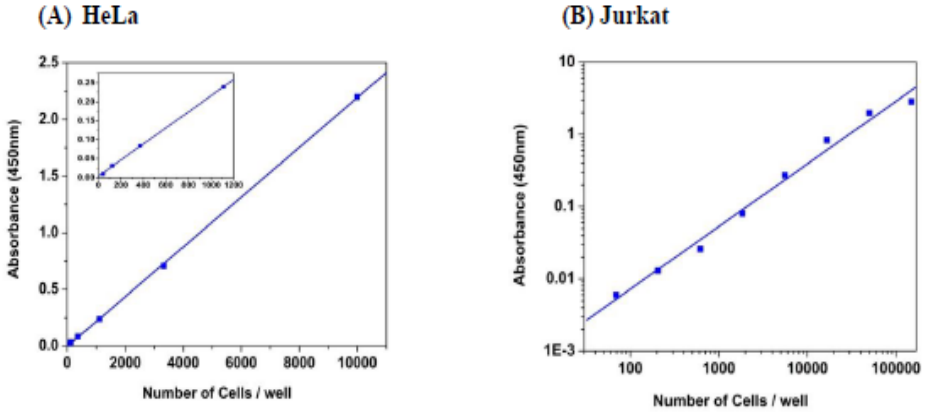

Figure 2: Cell number was determined with the cell Counting Kit 8 (WST-8) (ab228554). (A) HeLa cells at 0 to 10,000 cells/well/100 µL, and (B) Jurkat cells at 0 to 100,000 cells/well/100 µL were added in a clear bottom 96-well plate. The absorbance was measured at 460 nm using a plate reader.

Technical Support

For all technical or commercial enquiries please go to:
www.abcam.com/contactus
www.abcam.cn/contactus (China)
www.abcam.co.jp/contactus (Japan)