

**ab65473**

# **Quick Cell Proliferation Assay Kit**

## **Instructions for Use**

For the rapid, sensitive and accurate measurement of Cell Proliferation in cell culture (adherent and suspension)

This product is for research use only and is not intended for diagnostic use.

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# 1. Overview

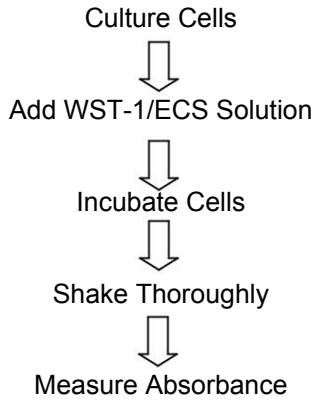
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Abcam's Quick Cell Proliferation Assay Kit provides all reagents and detailed instructions for a fast and sensitive quantification of cell proliferation and viability. The assay is based on the cleavage of the tetrazolium salt WST-1 to formazan by cellular mitochondrial dehydrogenases. Expansion in the number of viable cells resulted in an increase in the activity of the mitochondrial dehydrogenases, which leads to the increase in the amount of formazan dye formed. The formazan dye produced by viable cells can be quantified by multi-well spectrophotometer (microtiter plate reader) by measuring the absorbance of the dye solution at 440 nm.

The assay can be used for the measurement of cell proliferation in response to growth factors, cytokines, mitogens, and nutrients, etc. It can also be used for the analysis of cytotoxic compounds like anticancer drugs and many other toxic agents and pharmaceutical compounds. The new method is so simple, requiring no washing, no harvesting, and no solubilization steps, and is faster and more sensitive than MTT, XTT, or MTS-based assays. The entire assay can be performed in a microtiter plate.

## 2. Protocol Summary

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### 3. Components and Storage

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#### A. Kit Components

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Item	Quantity (500 assays)	Quantity (2500 assays)
WST Reagent IV	1 vial	1 vial
Electrocoupling Solution	5 ml	25 ml

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\* Store kit at -20°C.

WST Reagent IV: Dissolve 1 vial of lyophilized WST Reagent IV into 5 ml/25 ml Electrocoupling Solution aliquot the solution (1 ml is sufficient for one 96-well plate assay) and store at -20°C.

The WST Reagent IV / Electrocoupling Solution Solution is stable for 3 months at -20°C. Protect from light. Avoid repeated freeze-thaw. Repeated freeze-thaw may increase background.

## **B. Additional Materials Required**

- Microcentrifuge
- Pipettes and pipette tips
- Multi-well spectrophotometer (microtiter plate reader)
- 96 well plate
- Orbital shaker

## 4. Assay Protocol

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1. Culture cells ( $0.1\text{-}5 \times 10^4$  cells/well) in a 96-well microtiter plate in a final volume of 100  $\mu\text{l}$ /well culture medium in the absence or presence of various amounts of the factors tested.

For toxicity assays, use more cells to start with (e.g.  $5 \times 10^4\text{-} \times 10^5$  cells/well).

2. Incubate cells for 24-96 hours.
3. Add 10  $\mu\text{l}$  WST Reagent IV / Electrocoupling Solution solution to each well. Be careful not to introduce bubbles to the wells.

**Note:**

If the cells are cultured in different volumes of culture medium, increase or decrease the amount of WST Reagent IV / Electrocoupling Solution solution accordingly.

4. Incubate the cells for 0.5-4 hours in standard culture conditions.

**Note:**

The appropriate incubation time depends on the individual cell type and cell concentration used. Therefore, it is recommended to determine the optimal incubation time for the particular experimental setup used.

5. Shake thoroughly for 1 min.

6. Measure the absorbance of the treated and untreated samples using a microtiter plate reader at 420-480 nm according to the filters available for the plate reader. The reference wavelength should be ~650nm.

**Notes:**

- a) Use the same amount of culture medium and WST Reagent IV / Electrocoupling Solution solution in an empty well as a blank position for the microtiter plate reader.
- b) The assay can be stopped by adding 10  $\mu$ l of 1% SDS into each well, and shake to mix.
- c) Phenol Red in culture medium does not significantly interfere with the reading.



## Technical Support

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