

Product Information Sheet

## WST-1

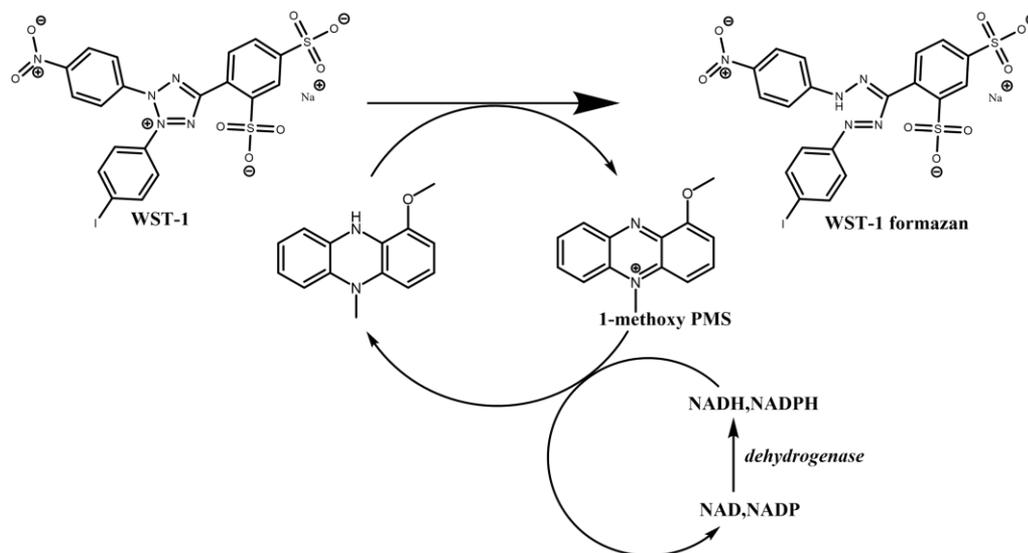
### Water-Soluble Tetrazolium 1

Catalogue Number SBR00125

### Product Description

WST-1 (water-soluble tetrazolium salt 1) is a potent reagent for evaluating cell proliferation, growth, and chemosensitivity in various cell populations.<sup>1-3</sup> This compound converts a stable tetrazolium salt into a soluble formazan dye at the cell surface, with dye production directly correlating to the number of viable, metabolically active cells.<sup>3</sup> To enable electron transfer from intracellular reducing agents to WST-1, an electron coupling reagent like 1-methoxy phenazine methosulfate (PMS) is often required, facilitating a colorimetric assay for cell viability. (Figure 1).

WST-1 is optimized for 96-well plate formats, allowing simultaneous processing of multiple samples. Its sensitivity makes it suitable for measuring cell proliferation in response to growth factors and nutrients,<sup>4-6</sup> as well as assessing the impact of growth-inhibitory antibodies and cytotoxic compounds, including anti-cancer drugs.<sup>7-9</sup> Additionally, WST-1 can monitor enzymatic reactions by utilizing NADH/NADPH electron transfer to PMS, applicable for measuring dehydrogenase activity or other electron transfer reactions.<sup>10</sup>



**Figure 1:** Cell viability detection mechanism of WST-1 with 1-methoxy PMS

## Reagents and Equipment Required but Not Provided

- Ultrapure water.
- 96 well flat-bottom plate
- Fluorescence multiwell plate reader
- 37 °C + 5%CO<sub>2</sub> incubator
- Multichannel pipettes (10 and 100 µl).
- 1-Methoxy PMS (Catalogue# M8640)
- HEPES (Catalogue# H0887)

## Precautions and Disclaimer

For R&D use only. Not for drug, household, or other uses. Please consult the Safety Data Sheet for information regarding hazards and safe handling practices.

## Storage/Stability

Store at -20 °C and protect from light.

## Preparation Instructions

WST-1 Working Solution Preparation for Cell Viability and Enzymatic Detection Assays:

Prepare WST-1 solution in an aqueous solution containing 5 mM WST-1, 0.2 mM 1-methoxy PMS (Catalogue# M8640), and 12.5 mM HEPES solution (Catalogue# H0887).

### Example Preparation (5 mL Solution):

1. Prepare a 12.5 mM HEPES stock solution. To a final volume of 20 mL water, add 250 µL of 1 M HEPES solution.
2. Prepare a stock solution of 2 mM of 3.4 mg 1-methoxy PMS in 5 mL buffer of 12.5 mM HEPES solution.
3. Dissolve 16.3 mg of WST-1 in 4 ml of 12.5 mM HEPES buffer. Then add 500 µL of the 2 mM 1-Methoxy PMS stock solution and mix. Bring to a final volume of 5 mL by adding 500 µL of 12.5mM HEPES stock solution.

## Recommended Storage of Solution

Store the solution at -20 °C. Under these conditions, the solution is stable for up to 2 years.

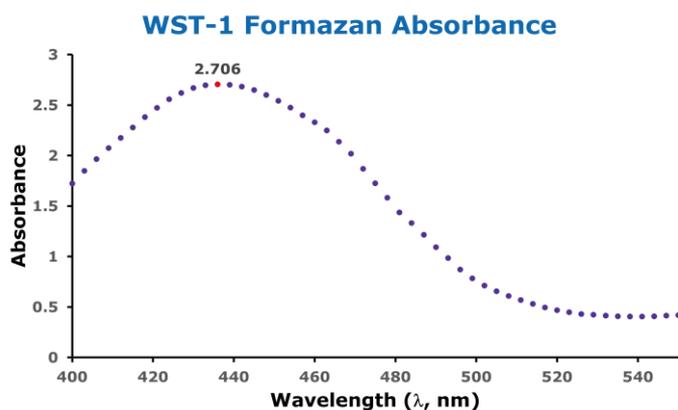
**Note:** To maintain the quality of the solution, avoid repeated freeze-thaw cycles. It is recommended to divide the solution into aliquots before freezing.

### Short-Term Storage:

The solution can be stored at 4 °C for approximately two months.

## Procedure for Cell Viability Assay

1. Grow cell suspension in a 96-well microplate (100 µl) at 37 °C in a humidified incubator with 5% CO<sub>2</sub> for the required time.
2. Add 10 µl of the WST-1 solution to the cells (1:10 dilution).
3. Incubate for 0.5-4 hours at 37 °C in a humidified incubator with 5% CO<sub>2</sub>
4. Measure the absorbance using a microplate reader at 420 to 480 nm (maximum absorption at ~440 nm, see Figure 2.)



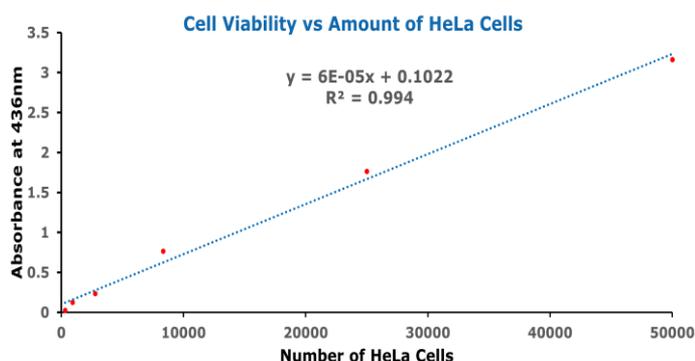
**Figure 2:** Well plate max wavelength ( $\lambda$ ) absorbance of WST-1 formazan analysis.

The graph above illustrates the maximum absorbance peak of WST-1 solution ( $\lambda_{\text{max}} = 436\text{nm}$ ) after incubation (2h) with viable cells, indicating the conversion of WST-1 into soluble formazan dye.

## Results

Based on the cell counting procedure above, WST-1 solution was tested on HELA cells ( $0.3\text{-}5 \times 10^4$ ) after 2 hours of incubation at  $37^\circ\text{C}$  humidified incubator with 5%  $\text{CO}_2$ . Microplate reading was performed at 436nm (see Figure 3).

**Note:** Microplate reading for WST-1 is commonly performed at 440-450 nm.



**Figure 3.**

WST-1 working solution cell viability vs. number of HeLa cells analysis.

## References

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