



Cell Technology, Inc

Superoxide Dismutase Assay

Key Benefits

- 100% Inhibition by Super Oxide Dismutase (SOD)
- Can detect low concentrations of SOD
- Highly water-soluble formazan dye.
- Applications: colorimetric detection.

Introduction

Superoxide dismutase (SOD) are metalloenzymes that catalyze the dismutation of superoxide radical into hydrogen peroxide (H₂O₂) + molecular oxygen (O₂) and consequently provide an important defense mechanism against superoxide radical toxicity (1).

Oxidative stress dependent upon superoxide radical can account for a number of acute and chronic disease states, which include inflammation and ischemia-reperfusion (2,3). SOD protects murine peritoneal macrophages from apoptosis induced by adriamycin (4). Furthermore, over expression of SOD in fibrosarcoma cells, protects against apoptosis and promotes cell differentiation (5).

Assay Principle

To determine SOD activity, several direct and indirect methods have been developed. A common and convenient indirect method utilizes nitroblue tetrazolium (NBT) conversion to NBT-diformazan (formazan dye) via superoxide radical. However, there are several disadvantages to the NBT method, such as poor water solubility of the formazan dye and the interaction with the reduced form of xanthine oxidase. Though cytochrome C is also commonly used for SOD activity detection, its reactivity with superoxide is too high to determine low levels of SOD activity.

Cell Technology's SOD kit utilizes a water-soluble tetrazolium salt, WST-1 (2-(4-Iodophenyl)-3-(4-nitrophenyl)-5-(2,4-disulfophenyl)-2H-tetrazolium, monosodium salt) that produces a highly water-soluble formazan dye upon reduction with a superoxide anion (6). The rate of the reduction with O₂⁻ is linearly related to the xanthine oxidase (XO) activity, and is inhibited by SOD, as shown in Figure 1. Therefore, the IC₅₀ (50% inhibition activity of SOD or SOD-like materials) can be determined by this colorimetric method. Absorbance can be measured at 440nm.

Figure 1 - Inhibition Curve Prepared Using SOD from Bovine Liver

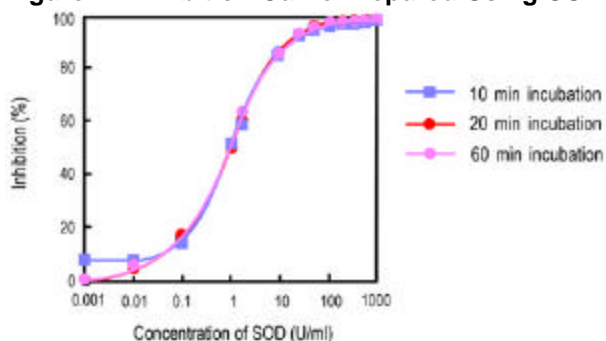
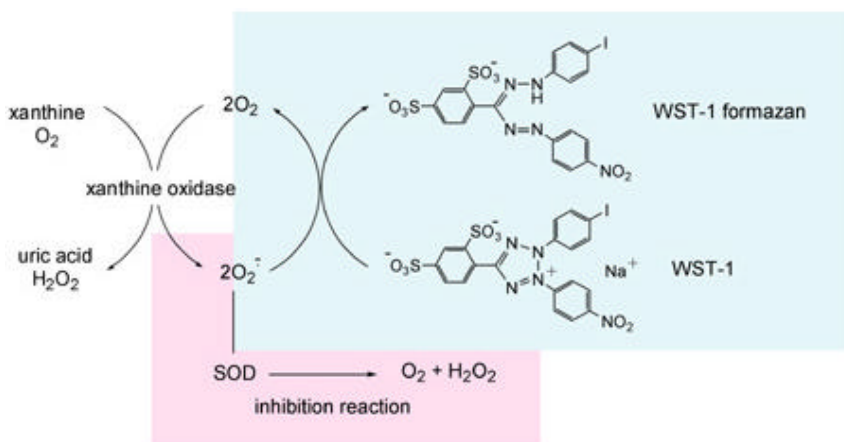


Figure 2 - SOD ASSAY Reaction:



Ordering Information

Catalog #	Contents	Size	Price (US\$)
CSOD100-2	See below*	100 Tests	225

*Kit Contents:

1. Part # 4014. 20X WST-1 Solution: 1 ml.
2. Part # 6010. Xanthine oxidase solution (XO): 20uL.
3. Part # 3029. Assay Buffer: 20 mL.
4. Part # 3030. Xanthine oxidase Dilution Buffer: 10mL. (XO dilution Buffer)
5. Part# 6011. SOD Enzyme: 30uL. See vial for activity.
6. Part# 9001. 96 well ELISA Plate: 1 plate
7. Part# 9002. Adhesive Plate Cover: 2.

References:

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2. Lontz, W., Sirsjo, J., Liu, W., Lindberg, M., Rollman, O., and G. (1976) *Int. J. Cancer* **17**, 62–70. Torma, H. (1995) *Free Radical Biol. Med.* **18**, 349–355.
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4. Dominguez-Rodriguez, J.R. et al. (2001) *Anticancer Res.* **21**:1869.
5. Zhao, Y. et al. (2001) *Antioxid. Redox Signal* **3**:375.
6. H. Ukeda, A. K. Sarker, D. Kawana and M. Sawamura, *Anal. Sci.*, **15**, 353 (1999).

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