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APB960Mu01 100µg Active Superoxide Dismutase 1 (SOD1) Organism Species: *Mus musculus (Mouse) Instruction manual*

FOR RESEARCH USE ONLY NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

1st Edition (Apr, 2016)

[PROPERTIES]

Source: Prokaryotic expression. Host: *E. coli* Residues: Ala2~Gln154 Tags: N-terminal His-tag Purity: >95% Endotoxin Level: <1.0EU per 1µg (determined by the LAL method). Buffer Formulation: PBS, pH7.4, containing 0.01% SKL, 5% Trehalose. Applications: Cell culture; Activity Assays. (May be suitable for use in other assays to be determined by the end user.) Predicted isoelectric point: 6.3 Predicted Molecular Mass: 17.0kDa Accurate Molecular Mass: 19kDa as determined by SDS-PAGE reducing conditions.

[<u>USAGE</u>]

Reconstitute in 10mM PBS (pH7.4) to a concentration of 0.1-1.0 mg/mL. Do not vortex.

[STORAGE AND STABILITY]

Storage: Avoid repeated freeze/thaw cycles.

Store at 2-8°C for one month.

Aliquot and store at -80°C for 12 months.

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Stability Test: The thermal stability is described by the loss rate. The loss rate was determined by accelerated thermal degradation test, that is, incubate the protein at 37°C for 48h, and no obvious degradation and precipitation were observed. The loss rate is less than 5% within the expiration date under appropriate storage condition.

[<u>SEQUENCE</u>]

AMKAVCVLK GDGPVQGTIH FEQKASGEPV VLSGQITGLT EGQHGFHVHQ YGDNTQGCTS AGPHFNPHSK KHGGPADEER HVGDLGNVTA GKDGVANVSI EDRVISLSGE HSIIGRTMVV HEKQDDLGKG GNEESTKTGN AGSRLACGVI GIAQ

[ACTIVITY]

Extracellular superoxide dismutase [Cu-Zn] is an enzyme that in humans is encoded by the SOD3 gene. This gene encodes a member of the superoxide dismutase (SOD) protein family. SODs are antioxidant enzymes that catalyze the dismutation of two superoxide radicals into hydrogen peroxide and oxygen. Acroding to the report, in a weakly alkaline buffer solution (pH=8.2) with N-tris(hydroxymethyl)amino methane-HCL, pyrogallol can occur autoxidation in the air, then SOD can inhibit this reaction. Thus, we use this way to measued the activity of recombinant mouse SOD1. The reaction was performed in adding 15 µ I 5mmol/L pyrogallol to 900 µ I 50mmol/L Tris-HCl in 1.5ml cuvette(1.0 cm light path), rapidly mixing at 25 °C, then read at 325nm (zero the spectrophotometer using 50mmol/L Tris-HCI), reacod the OD value every 30 second for 6 times. Control the pyrogallol autoxidation rate at 0.70 OD/min. After, adding various concentrations of recombinant of SOD1 to 900 µ I 50mmol/L Tris-HCI, incubated for 20min at 25 °C, then adding 15µl 5mmol/L pyrogallol to each tube, rapidly mixing and read at 325nm, reacod the OD value every 30 second for 6 times. Under these conditions, the enzyme amount of 50% per minute inhibition of pyrogallol autooxidation is defined as a unit.



Calculation

SOD1 activity (U/mI) =
$$\frac{\frac{0.070 - A325 / \min}{0.070} \times 100\%}{50\%} \times 0.915 / V \times N$$

Where:

0.070=pyrogallol autoxidation rate

A325/min= inhibition pyrogallol autoxidation rate of SOD1

0.915= total volume in cuvette

V=sample volume

N=dilution factor

Specific SOD1 activity (U/mg)=SOD1 activity/amount of SOD1in 1ml.

The specific activity of recombinant mouse SOD1 is 374U/mg

[IDENTIFICATION]

kDa 70
44
33
26
22
18
14
10

Figure 1. SDS-PAGE

Sample: Active recombinant SOD1, Mouse

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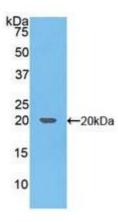


Figure 2. Western Blot

Sample: Recombinant SOD1, Mouse;

Antibody: Rabbit Anti- Mouse SOD1 Ab (PA B960Mu01)

[IMPORTANT NOTE]

The kit is designed for research use only, we will not be responsible for any issue if the kit was used in clinical diagnostic or any other procedures.