APA117Hu01 200µg Active Superoxide Dismutase 3, Extracellular (SOD3) Organism Species: *Homo sapiens (Human) Instruction manual*

FOR RESEARCH USE ONLY NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

13th Edition (Revised in Aug, 2023)

[PROPERTIES]

Source: Prokaryotic expression. Host: *E. coli* Residues: Trp19~Ala240 Tags: N-terminal His-tag Purity: >90% Endotoxin Level: <1.0EU per 1µg (determined by the LAL method). Buffer Formulation: PBS, pH7.4, containing 0.01% SKL, 5% Trehalose. Original Concentration: 800µg/mL 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: 27.9kDa Accurate Molecular Mass: 33/30kDa 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.

C Cloud-Clone Corp. 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.

[SEQUENCE]

WT GEDSAEPNSD SAEWIRDMYA KVTEIWQEVM QRRDDDGALH AACQVQPSAT LDAAQPRVTG VVLFRQLAPR AKLDAFFALE GFPTEPNSSS RAIHVHQFGD LSQGCESTGP HYNPLAVPHP QHPGDFGNFA VRDGSLWRYR AGLAASLAGP HSIVGRAVVV HAGEDDLGRG GNQASVENGN AGRRLACCVV GVCGPGLWER QAREHSERKK RRRESECKAA

[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. According 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 measure the activity of recombinant human SOD3. The reaction was performed in adding 30µL 50mmol/L pyrogallol to 900µL 50mmol/L Tris-HCl in 1.5mL cuvette (1.0cm light path), rapidly mixing at 25°C, then read at 325nm (zero the spectrophotometer using 50mmol/L Tris-HCI), record 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 SOD3 to 900µL 50mmol/L Tris-HCl, incubated for 20min at 25°C, then adding 30µL 50mmol/L pyrogallol to each tube, rapidly mixing and read at 325nm, record 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.

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Calculation:

$$\frac{0.070 - A325 / \min}{0.070} \times 100\%$$
SOD3 activity (U/ml) = 50%
Where:
0.070=pyrogallol autoxidation rate
A325/min=inhibition pyrogallol autoxidation rate of SOD3
0.93=total volume in cuvette
V=sample volume
N=dilution factor
Specific SOD3 activity (U/mg)=SOD3 activity/amount of SOD3 in 1mL
The specific activity of recombinant human SOD3 is 121.4U/mg

[IDENTIFICATION]

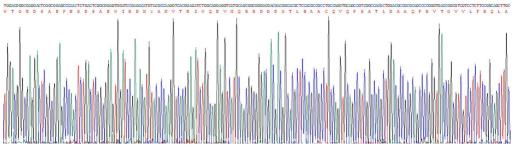


Figure 1. Gene Sequencing (extract)

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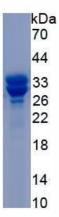


Figure 2. SDS-PAGE

Sample: Active recombinant SOD3, Human

[<u>IMPORTANT NOTE</u>]

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.