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APC418Hu05 100µg Active Catalase (CAT) Organism Species: *Homo sapiens (Human) 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: Asp10~Asn507 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. Applications: Cell culture; Activity Assays. (May be suitable for use in other assays to be determined by the end user.) Predicted isoelectric point: 6.9 Predicted Molecular Mass: 60.4kDa Accurate Molecular Mass: 60kDa 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.

[SEQUENCE]

D QMQHWKEQRA AQKADVLTTG AGNPVGDKLN VITVGPRGPL LVQDVVFTDE MAHFDRERIP ERVVHAKGAG AFGYFEVTHD ITKYSKAKVF EHIGKKTPIA VRFSTVAGES GSADTVRDPR GFAVKFYTED GNWDLVGNNT PIFFIRDPIL FPSFIHSQKR NPQTHLKDPD MVWDFWSLRP ESLHQVSFLF SDRGIPDGHR HMNGYGSHTF KLVNANGEAV YCKFHYKTDQ GIKNLSVEDA ARLSQEDPDY GIRDLFNAIA TGKYPSWTFY IQVMTFNQAE TFPFNPFDLT KVWPHKDYPL IPVGKLVLNR NPVNYFAEVE QIAFDPSNMP PGIEASPDKM LQGRLFAYPD THRHRLGPNY LHIPVNCPYR ARVANYQRDG PMCMQDNQGG APNYYPNSFG APEQQPSALE HSIQYSGEVR RFNTANDDNV TQVRAFYVNV LNEEQRKRLC ENIAGHLKDA QIFIQKKAVK NFTEVHPDYG SHIQALLDKY NAEKPKN

[ACTIVITY]

Catalase (CAT) is an antioxidant enzyme present in all aerobic organisms. It is known to catalyze H_2O_2 into water and oxygen in an energy-efficient manner in the cells exposed to environmental stress. As we know, H2O2 and (NH4)2MoO4 can produce a stable yellow product which has a absorbance at 405 nm. Thus, the recombinant human CAT activity was measured by its ability to hydrolyze H_2O_2 to H_2O , the remaining H_2O_2 in the reaction was measured by (NH4)2MoO4. The reaction was performed in 60 mM phosphate buffer pH 7.4 (Assay Buffer), initiated by addition 100 µL of various concentrations of CAT (diluted by Assay Buffer) to 1 mL of 65 mM Substrate H_2O_2 (diluted by Assay Buffer). Incubated at 37°C for 1min, then add 1 ml 200 mM (NH4)2MoO4. After 10 minutes at room temperature, read at a wavelength of 405 nm.

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Concentration of H ₂ O ₂ (mM)	OD (405nm)
32.5	1.325
16.25	0.604
8.125	0.311
4.0625	0.181
2.03125	0.078
1.015625	0.038
0.5078125	0.013

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Figure 1. The standard curve of H₂O₂

One unit of enzyme activity is defined as the 1µg of enzyme required to convert 1umol of H₂O₂ to H₂O in 1min at 37°C. The specific activity of recombinant human CAT is 53.8 umol/min/µg.

[IDENTIFICATION]



Figure 2. Gene Sequencing (extract)

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kDa 70
44
33
26
22
18
14
 10

Figure 3. SDS-PAGE

Sample: Active recombinant CAT, Human



Figure 4. Western Blot

Sample: Recombinant CAT, Human;

Antibody: Rabbit Anti-Human CAT Ab (PAC418Hu05)

[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.