

**APC418Ra01 100µg
Active Catalase (CAT)**

**Organism Species: *Rattus norvegicus* (Rat)
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: Asp3~Leu527

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: 7.5

Predicted Molecular Mass: 63.2kDa

Accurate Molecular Mass: 64kDa as determined by SDS-PAGE reducing conditions.

[USAGE]

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.

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]

DSRDPASDQMKQWKEQRAPQKPDVLTGGGNPIGDKLNIIMTAGPRGPLLVDVVFTDEMAHFDRERIPERVVHAKGAGAFGYFEVTHD
 ITRYSKAKVFEHIGKRTPIAVRFSTVAGESGADTVRDRGFAVKFYTEDGNWDLVGNNTPIFFIRDAMLFPFSIHSQKRNPQTHLKD
 PDMVWDFWSLCPESLHQVTFLFSDRGIPDGRHMRMNGYGSHTFKLVNANGEAVYCKFHYKTDQGIKNLPVEEAGRLAQEDPDYGLRDLF
 NAIASGNYPSTWTFYIQVMTFKEAETFPNPFDLTKVWPHKDYPLIPVGKLVLRNRPANYFAEVEQMAFDPSNMPPGIEPSPDKMLQGR
 LFAYPDTHRHRLGPNYLQIPVNCPRARVANYQRDGPMMHDNQGGAPNYYPNSFSAPEQQGSALEHHSQCSADVKRFN SANEDNVTQ
 VRTFYTKVLNEEERKRCENIANHLKDAQLFIQKAVKNFTDVHPDYGARVQALLDQYNSQKPKNAIHTYVQAGSHIAAKGKANL

[ACTIVITY]

Catalase (CAT) is an antioxidant enzyme present in all aerobic organisms. It is known to catalyze H₂O₂ into water and oxygen in an energy-efficient manner in the cells exposed to environmental stress. H₂O₂ will have specific absorbance at 240 nm . when we add CAT, the absorbance will decrease, thus the activity of CAT can be measured by calculating H₂O₂ absorbance decrease. The reaction was performed in adding 10ul (dilute with 50mM Potassium Phosphate Buffer, pH 7.0) recombinant Rat CAT to 290ul substrate mixture solution (50mM Potassium Phosphate Buffer, pH 7.0, 0.036% H₂O₂, allow the substrate to equilibrate to 25 °C), quickly mixed, then record the time required for the A240 to decrease from 0.45 to 0.40 absorbance units. One unit of catalase will decompose 1.0 μmole of H₂O₂ per minute at pH 7.0 at 25 °C. The activity of recombinant Rat CAT is 176000 U/mg.

Calculation

$$\text{CAT(U/mg)} = \frac{3.45 * d}{\text{time}} * 0.1 / \text{mg enzymatic}$$

Where:

3.45=corresponds to the decomposition of 3.45 μ moles of hydrogen peroxide in a 3.0 ml reaction mixture producing a decrease in the A240 from 0.45 to 0.40

d=dilution factor

Time=minutes required for the A240 to decrease from 0.45 to 0.40 absorbance units

0.1 = milliliter of enzyme added to the cuvette

[**IDENTIFICATION**]

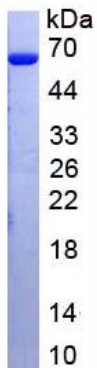


Figure 3. SDS-PAGE

Sample: Active recombinant CAT, Rat

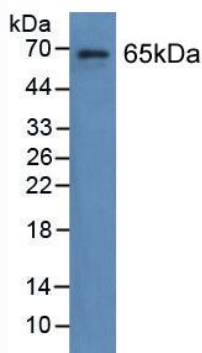


Figure 4. Western Blot

Sample: Recombinant CAT, Rat;

Antibody: Rabbit Anti- Rat CAT Ab (PAC418Ra01)

[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.