APC270Hu01 100µg Active 7-Dehydrocholesterol Reductase (DHCR7) 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: Cys61~Leu153 Tags: N-terminal His and GST 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 . Original Concentration: 200µg/mL Applications: Cell culture; Activity Assays. (May be suitable for use in other assays to be determined by the end user.) Predicted isoelectric point: 7.9 Predicted Molecular Mass: 40.2kDa Accurate Molecular Mass: 41kDa 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]

CDQYSCALTG PVVDIVTGHA RLSDIWAKTP PITRKAAQLY TLWVTFQVLL YTSLPDFCHK FLPGYVGGIQ EGAVTPAGVV NKYQINGLQA WLL

[ACTIVITY]

DHCR7 (7-dehydrocholesterol reductase) is a rate-limiting enzyme in cholesterol synthesis, which is mainly involved in the conversion of 7-dehydrocholesterol (7-DHC) to cholesterol. In the cholesterol biosynthetic pathway, this protease can reduce the C7-C8 double bond of cholesta-5,7-dien-3beta-ol (7-dehydrocholesterol/7-DHC) and cholesta-5,7,24-trien-3beta-ol, two intermediates in that pathway. In the absence of functional DHCR7, accumulation of 7DHC and a lack of cholesterol production leads to the devastating developmental disorder, Smith-Lemli-Opitz syndrome. Cholesterol 25-hydroxylase (CH25H) can also repress cholesterol biosynthetic enzymes. A functional ELISA assay was conducted to detect the interaction of recombinant human DHCR7 and recombinant human CH25H. Briefly, biotin-linked DHCR7 were diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of 100 ul were then transferred to CH25H-coated microtiter wells and incubated for 1h at 37°C. Wells were washed with PBST 3 times and incubation with Streptavidin-HRP for 30 min, then wells were aspirated and washed 5 times. With the addition of substrate solution, wells were incubated 15-25 minutes at 37 °C. Finally, add 50 µl stop solution to the wells and read at 450 nm immediately. The binding activity of recombinant human

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DHCR7 and recombinant human CH25H was shown in Figure 1, the EC50 for this effect is 0.028 ug/mL.



Figure 1. The binding activity of recombinant human DHCR7 and recombinant human CH25H

[IDENTIFICATION]



Figure 2. Gene Sequencing (extract)

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Figure 3. SDS-PAGE

Sample: Active recombinant DHCR7, 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.