APA656Hu61 100μg
Active Diamine Oxidase (DAO)
Organism Species: *Homo sapiens* **(Human)**
Instruction manual

FOR RESEARCH USE ONLY
NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

1th Edition (Apr, 2016)

[PROPERTIES]

Source: Eukaryotic expression.

Host: 293F cell

Residues: Glu20~Val751

Tags: N-terminal His-tag

Purity: >98%

Endotoxin Level: <1.0EU per 1μg (determined by the LAL method).

Buffer Formulation: 10mM PBS, pH7.6, containing 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: 6.6

Predicted Molecular Mass: 85.1kDa

Accurate Molecular Mass: 100kDa as determined by SDS-PAGE reducing conditions.

Phenomenon explanation:

The possible reasons that the actual band size differs from the predicted are as follows:

1. Splice variants: Alternative splicing may create different sized proteins from the same gene.

2. Relative charge: The composition of amino acids may affects the charge of the protein.

3. Post-translational modification: Phosphorylation, glycosylation, methylation etc.

4. Post-translation cleavage: Many proteins are synthesized as pro-proteins, and then cleaved to give the active form.

5. Polymerization of the target protein: Dimerization, multimerization etc.

[USAGE]

Reconstitute in 10mM PBS (pH7.6) 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]

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              E PSPGTLPRKA GVFSDLSNQE LKAVHSFLWS
KKELRLQPSS TTTMAKNTVF LIEMLLPKKY HVLRFLDKGE RHPVREARAV
IFFGDQEHPN VTEFAVGPLP GPCYMRALSP RPGYQSSWAS RPISTAEYAL
LYHTLQEATK PLHQFFLNTT GFSFQDCHDR CLAFTDVAPR GVASGQRRSW
LIIQRYVEGY FLHPTGLELL VDHGSTDAGH WAVEQVWYNG KFYGSPEELA
RKYADGEVDV VVLEDPLPGG KGHDSTEEPP LFSSHKPRGD FPSPIHVSGP
RLVQPHGPRF RLEGNAVLYG GWSFAFRLRS SSGLQVLNVH FGGERIAYEV
SVQEAVALYG GHTPAGMQTK YLDVGWGLGS VTHELAPGID CPETATFLDT
FHYYDADDPV HYPRALCLFE MPTGVPLRRH FNSNFKGGFN FYAGLKGQVL
VLRTTSTVYN YDYIWDFIFY PNGVMEAKMH ATGYVHATFY TPEGLRHGTR
LHTHLIGNIH THLVHYRVDL DVAGTKNSFQ TLQMKLENIT NPWSPRHRVV
QPTLEQTQYS WERQAAFRFK RKLPKYLLFT SPQENPWGHK RTYRLQIHSM
ADQVLPPGWQ EEQAITWARY PLAVTKYRES ELCSSSIYHQ NDPWHPPVVF
EQFLHNNENI ENEDLVAWVT VGFLHIPHSE DIPNTATPGN SVGFLLRPFN
FFPEDPSLAS RDTVIVWPRD NGPNYVQRWI PEDRDCSMPP PFSYNGTYRP
V
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[ACTIVITY]

Diamine oxidase (DAO), also known as histaminase, is an enzyme involved in the metabolism, oxidation, and inactivation of histamine in animals. Highest content is observed in the digestive tract and placenta. It is also secreted by eosinophils. In case of a shortage of diamine oxidase in the human body, it may appear as an allergy or histamine intolerance. Besides, Dopa Decarboxylase (DDC) has been identified as an interactor of DAO, thus a binding ELISA assay was conducted to detect the interaction of recombinant human DAO and recombinant human DDC. Briefly, DAO were diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of 100μL were then transferred to DDC-coated microtiter wells and incubated for 2h at 37℃. Wells were washed with PBST and incubated for 1h with anti-DAO pAb, then aspirated and washed 3 times. After incubation with HRP labelled secondary antibody, wells were aspirated and washed 3 times. With the addition of substrate solution, wells were incubated 15-25 minutes at 37℃. Finally, add 50μL stop solution to the wells and read at 450nm immediately. The binding activity of DAO and DDC was shown in Figure 1, and this effect was in a dose dependent manner.
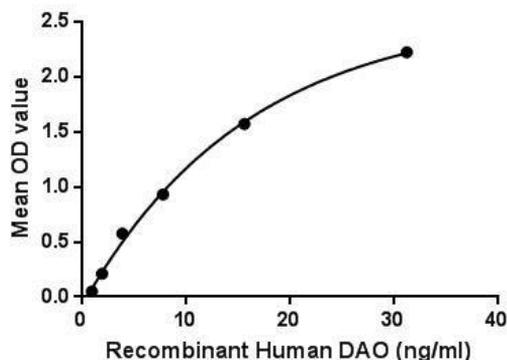

Figure 1. The binding activity of DAO with DDC.

[IDENTIFICATION]

EPSPGTLPRKAGVFSDLSNQELKAVHSFLWSKKELRLQPSSTTTMAKNTVFLIEMLLPKKYHVLRFLDKGER

Figure 2. Gene Sequencing (extract)

Figure 3. SDS-PAGE

Sample: Active recombinant DAO, Human

[IMPORTANT NOTE]

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