

APG963Hu01 100µg

Active Thymidylate Synthetase (TYMS)

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: Gln36~Val313

Tags: N-terminal His-tag

Purity: >93%

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

Buffer Formulation: 20mM Tris, 150mM NaCl, pH8.0, containing 0.05% sarcosyl and 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: 6.8

Predicted Molecular Mass: 35.6kDa

Accurate Molecular Mass: 36kDa as determined by SDS-PAGE reducing conditions.

[USAGE]

Reconstitute in 20mM Tris, 150mM NaCl (pH8.0) 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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                                                                 QIQHI LRCGVRKDDR
TGTGTLVFG MQARYSLRDE FPLLTKRVF WKGVLEELLW FIKGSTNAKE
LSSKGVKIWD ANGSRDFLDS LGFSTREEGD LGPVYGFQWR HFGAEYRDME
SDYSGQGVDQ LQRVIDTIKT NPDDRIIMC AWNPRDLPLM ALPPCHALCQ
FYVVNSELSC QLYQRSGDMG LGVPFNIAZY ALLTYMIAHI TGLKPGDFIH
TLGDAHIYLN HIEPLKIQLQ REPRPFKLR ILRKVEKIDD FKAEDFQIEG
YNPHPTIKME MAV
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[ACTIVITY]

Thymidylate synthetase (TYMS) is an enzyme that catalyzes the conversion of deoxyuridine monophosphate (dUMP) to deoxythymidine monophosphate (dTMP). The enzyme is essential for regulating the balanced supply of the 4 DNA precursors in normal DNA replication: defects in the enzyme activity affecting the regulation process cause various biological and genetic abnormalities. Thymidylate synthase is an enzyme of about 30 to 35kDa in most species except in protozoan and plants where it exists as a bifunctional enzyme that includes a dihydrofolate reductase domain. Besides, Aspartate Aminotransferase (AST) has been identified as an interactor of TYMS, thus a binding ELISA assay was conducted to detect the interaction of recombinant human TYMS and recombinant human AST. Briefly, TYMS were diluted serially in PBS with 0.01% BSA (pH 7.4). Duplicate samples of 100uL were then transferred to AST-coated microtiter wells and incubated for 2h at 37°C. Wells were washed with PBST and incubated for 1h with anti-TYMS 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 °C . Finally, add 50µL stop solution to the wells and read at 450nm immediately. The binding activity of TYMS and AST was shown in Figure 1, and this effect was in a dose dependent manner.

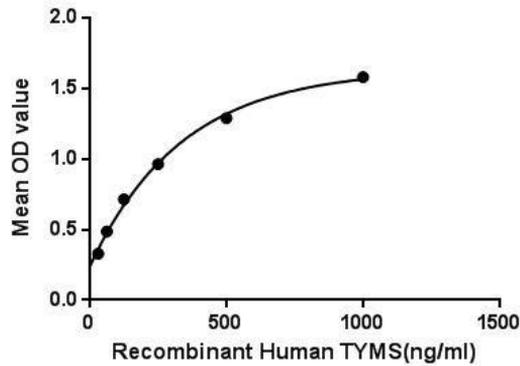


Figure 1. The binding activity of TYMS with AST.

[IDENTIFICATION]

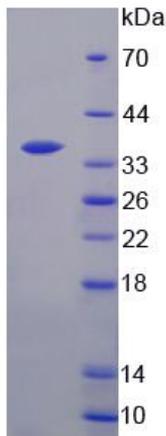


Figure 2. SDS-PAGE

Sample: Active recombinant TYMS, Human

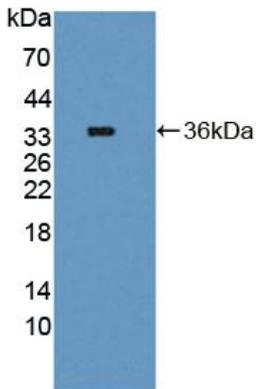


Figure 3. Western Blot

Sample: Recombinant TYMS, Human;

Antibody: Rabbit Anti-Human TYMS Ab (PAG963Hu01)

[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.