

**APG790Mu01 100µg**  
**Active Mdm2 p53 Binding Protein Homolog (MDM2)**  
**Organism Species: *Mus musculus (Mouse)***  
***Instruction manual***

FOR RESEARCH USE ONLY  
NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

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13th Edition (Revised in Aug, 2023)

## **[ PROPERTIES ]**

**Source:** Prokaryotic expression.

**Host:** *E. coli*

**Residues:** Met1~Asn310

**Tags:** N-terminal His-tag

**Purity:** >95%

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

**Buffer Formulation:** 20mM Tris, 150mM NaCl, pH8.0, containing 1mM EDTA, 1mM DTT, 0.01% SKL, 5% Trehalose and Proclin300.

**Original Concentration:** 200µg/mL

**Applications:** Activity Assays.

(May be suitable for use in other assays to be determined by the end user.)

**Predicted isoelectric point:** 4.2

**Predicted Molecular Mass:** 38.2kDa

**Accurate Molecular Mass:** 48kDa 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.

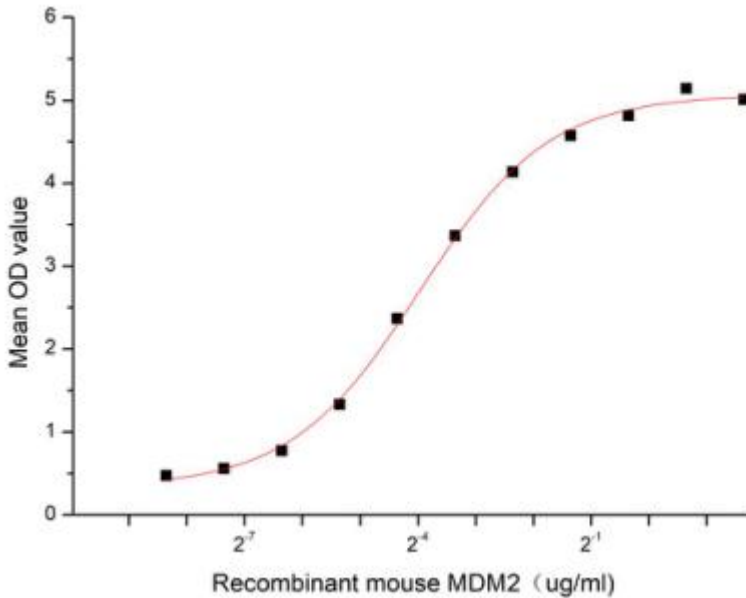
## [ **SEQUENCE** ]

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MCNTNMSVST EGAASTSQIP ASEQETLVRP KPLLLKLLKS VQAQNPTYTM
KEIIFYIGQY IMTKRLYDEK QQHIVYCSND LLGDVFGVPS FSVKEHRKIY
AMIYRNLVAV SQQDSGTSL SERRQPEGGS DLKDPLQAPP EEKPSDDL I
SRLSTSSRRR SISETEENTD ELPGERHRKR RRSLSFDPSL GLCELREMCS
GGSSSSSSSS SESETPSHQ DLDDGVSEHS GDCLDQDSVS DQFSVEFEVE
SLDSEDYSL SDEGHELSDED DEVYRVTVYQ TGESDTSFE GDPEISLADY
WKCTSCNEMN
```

## [ **ACTIVITY** ]

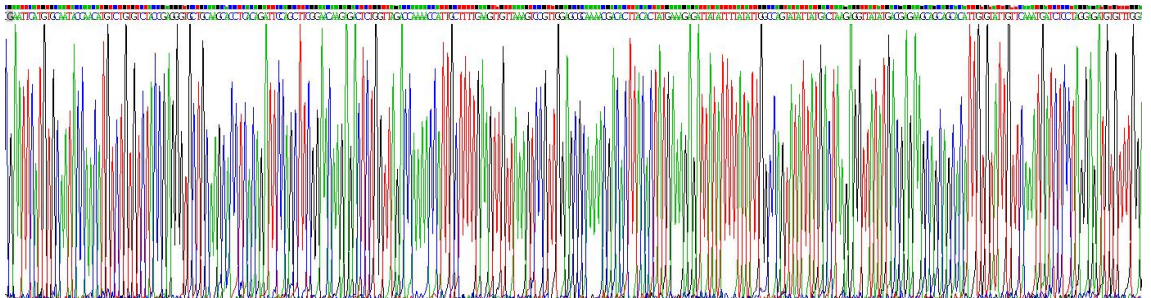
Mouse double minute 2 homolog (MDM2) also known as E3 ubiquitin-protein ligase. Mdm2 is a cellular oncoprotein that recognizes the N-terminal trans-activation domain (TAD) of the p53 tumor suppressor and as an inhibitor of p53 transcriptional activation. The human homologue of this protein is sometimes called Hdm2. The p53 tumor suppressor is the key target of MDM2. It has been identified as a p53 interacting protein that represses p53 transcriptional activity and also acts as an E3 ubiquitin ligase, targeting both itself and p53 for degradation by the proteasome. MDM2 is capable of auto-polyubiquitination, and in complex with p300, a cooperating E3 ubiquitin ligase, is capable of polyubiquitinating p53. Thus a binding ELISA assay was conducted to detect the interaction of recombinant mouse MDM2 and recombinant human P53. Briefly, biotin-linked MDM2 were diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of 100  $\mu$ l were then transferred to P53-coated microtiter wells and incubated for 1h at 37 °C . Wells were washed with PBST 3 times and incubation with Streptavidin-HRP for 30min, 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 50µl stop solution to the wells and read at 450nm immediately. The binding activity of MDM2 and P53 was shown in Figure 1, the EC50 for this effect is 0.064ug/mL.

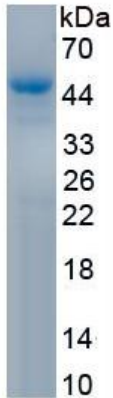


**Figure 1. The binding activity of recombinant mouse MDM2 and recombinant human P53**

**[ IDENTIFICATION ]**



**Figure 2. Gene Sequencing (extract)**



**Figure 3. SDS-PAGE**

**Sample: Active recombinant MDM2, Mouse**

**[ 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.