

APA853Mu61 100µg

Active Caspase 8 (CASP8)

Organism Species: *Mus musculus* (Mouse)

Instruction manual

FOR RESEARCH USE ONLY

NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

13th Edition (Revised in Aug, 2023)

[PROPERTIES]

Source: Eukaryotic expression.

Host: 293F cell

Residues: Ser219~Gly376

Tags: N-terminal His-tag

Purity: >90%

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

Buffer Formulation: PBS, pH7.4, containing 5% Trehalose .

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: 7.0

Predicted Molecular Mass: 19.4kDa

Accurate Molecular Mass: 20kDa 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]

SESRTSDKVYQMKNKPRGYCLIINNHDFSKAREDTQLRKMKDRKGTDCDKEALSKTFKEL
HFEIVSYDDCTANEIHEILEGYQSADHKNKDCFICCILSHGDKGVVYGTGKEASIYDLTSY
FTGSKCPSLSGKPKIFFIQACQGSNFQKGVPEAG

[ACTIVITY]

Caspase 8 (CASP8) is a key initiator protease in the extrinsic apoptosis pathway. Upon activation by death receptors—such as Fas or TNFR1—it is recruited to death-inducing signaling complexes (DISCs). Via its N-terminal death effector domains (DEDs), CASP8 forms oligomers and undergoes autocleavage to achieve full enzymatic activity; it then cleaves downstream effector caspases to execute the apoptotic program. Beyond apoptosis, CASP8 also plays a role in regulating necroptosis, inflammatory responses, and cell differentiation processes. Mechanistically, CASP8 interacts with TNFRSF1A Associated Via Death Domain Protein (TRADD) through homotypic death domain (DD)-mediated binding, a process that facilitates the transduction of apoptotic signals. To detect the activity of recombinant CASP8, a functional ELISA assay was performed to evaluate the interaction between recombinant mouse CASP8 and recombinant human TRADD. Briefly, biotin-linked CASP8 were diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of 100µl were then transferred to TRADD-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 CASP8 and TRADD was shown in Figure 1, the EC50 for this effect is 0.51µg/mL.

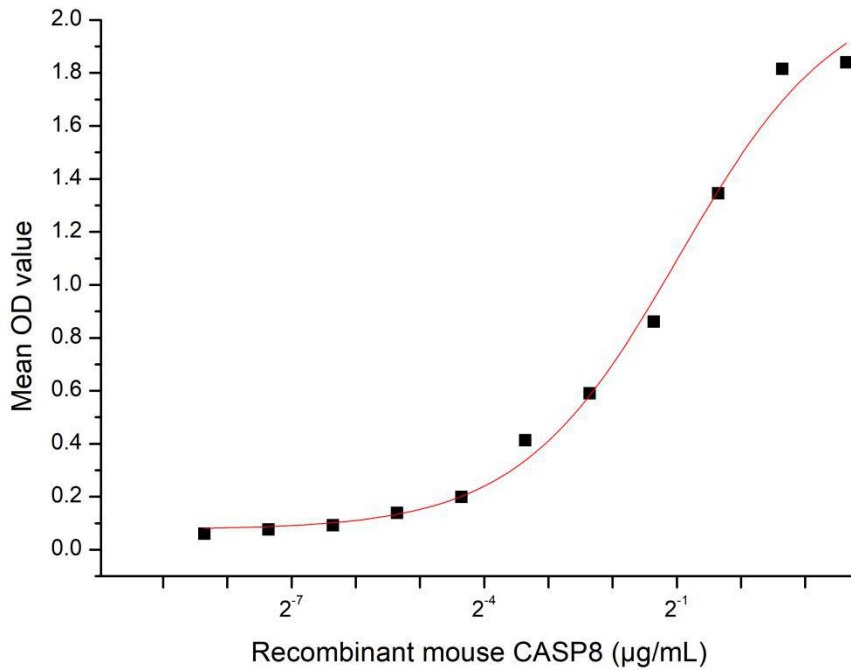


Figure 1. The binding activity of recombinant mouse CASP8 and recombinant human TRADD

[IDENTIFICATION]

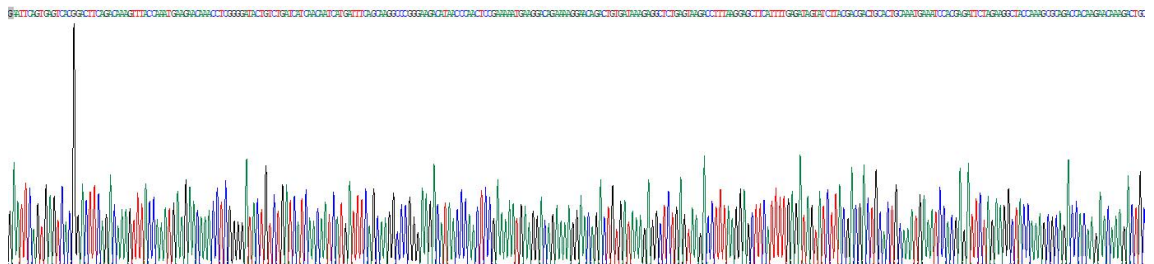


Figure 2. Gene Sequencing (extract)

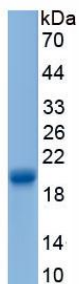


Figure 3. SDS-PAGE

Sample: Active recombinant CASP8, 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.