

APB209Mu01 100µg
Active Granzyme K (GZMK)
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: Prokaryotic expression.

Host: *E. coli*

Residues: Gln44~Val227

Tags: N-terminal His-tag

Purity: >90%

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: 9.6

Predicted Molecular Mass: 24.1kDa

Accurate Molecular Mass: 24kDa 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]

QYRSKHI
CGGVLIHPQW VLTAACHCYSW FPRGHSPTVV LGAHLSKNE PMKQTFEIKK
FIPF SRLQSG SASHDIMLIK LRTAAELNKN VQLLHLGSKN YLRDGTKCQV
TGWGTTKPD L LTASDTLREV TVTIISRKRC NSQSYNHKP VITKDMICAG
DARGQKDSCK GDSGGPLICK GIFHALV

[ACTIVITY]

Granzyme K is a member of the granzyme family of the serine proteases found specifically in the cytotoxic granules of cytotoxic T lymphocytes (CTL) and natural killer (NK) cells. Mouse granzyme K is synthesized as a precursor (263 residues) with a signal peptide (residues 1-21), a propeptide (residues 22-25) and a mature chain (residues 26-263). The activity of recombinant mouse Granzyme K was measured by its ability to cleaves a thioester substrate Z-Lys-SBzl • HCl. The reaction was performed in 0.05 M Tris, 0.15 M NaCl, 0.01% Triton X-100, pH 8.0 (assay buffer), initiated by addition 50 μ L of various concentrations of GZMK (diluted by assay buffer) to 50 μL of 1.2 mM substrate and DTNB mixture. The final well serves as a negative control with no GZMK, replace with 50 μ L assay buffer. Incubated at 25 °C for 5min, then read at a wavelength of 405 nm. The specific activity of recombinant mouse Granzyme K is >70 pmol/min/μg.

Specific Activity (pmol/min/ug)=

$\frac{\text{Adjusted } V_{\max}^* \text{ (OD/min)} \times \text{well volume (L)} \times 10^{12} \text{ pmol/mol}}{\text{ext. coeff}^{**} \text{ (M}^{-1}\text{cm}^{-1}) \times \text{path corr.}^{***} \text{ (cm)} \times \text{amount of enzyme (ug)}}$

*Adjusted for Substrate Blank

**Using the extinction coefficient 13800 M⁻¹cm⁻¹

***Using the path correction 0.320 cm

[IDENTIFICATION]

GAATTCCAGTACCCGACGCMGCATATTTGTGGAGGAGTCTGATCCACCCGACAGTGGGTGCTAACAGCGCGCCCTCTCTACTCTTGGTTCCCGA GAGGDCACTCTCCACCGTGGTTTTTA GGAGGACATTCTCTTTCCAGGATGAGCCCATGAAQAGACATTTGAAATTA AAAA GTTC

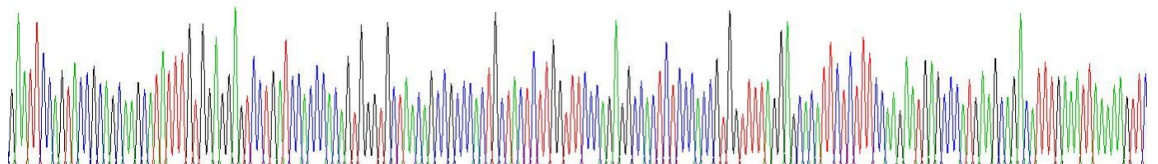


Figure 1. Gene Sequencing (extract)

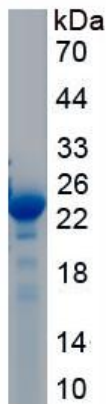


Figure 2. SDS-PAGE

Sample: Active recombinant GZMK, 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.