

APA600Po01 100µg
Active Granzyme B (GZMB)
Organism Species: *Sus scrofa*; *Porcine (Pig)*
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: Gly19~Leu246

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: 10.2

Predicted Molecular Mass: 29.2kDa

Accurate Molecular Mass: 29kDa 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]

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GEIIGGHEAKPHSRPYMAYLQYWNQDVQSRCGGFLVRQDFVLTAAHCNGSSIKVTLGAHN  
IKQQERTQQVIRVRRRAISHPDYNPKNFSNDIMLLKLERKAKQTSAVKPLSLPRAKARVKP  
GQTCVAGWGRDSTDYADTLQEVKLIQEDQKCEAYLRNFYNRAIQLCVGDPKTKKASF  
QGDSGGPLVCDNVAQGI VSYGKRDGSTPRAFTKVSSFLPWIKKTKMSL
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[ACTIVITY]

Granzyme B 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. Granzyme B plays an essential role in granule-mediated apoptosis and may have additional roles in rheumatoid arthritis and in bacterial and viral infections. It activates various caspases and cleaves proteins such as aggrecan. The activity of recombinant pig Granzyme B 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 GZMB (diluted by Assay Buffer) to 50 μ L of 1.2 mM Substrate and DTNB mixture. The final well serves as a negative control with no GZMB, replaced 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 pig Granzyme B is >440 pmol/min/ μ g.

Specific Activity (pmol/min/ug)=

Adjusted V_{max}^* (OD/min) x well volume (L) x 10^{12} pmol/mol

ext. coeff** ($M^{-1}cm^{-1}$) x path corr.*** (cm) x amount of enzyme (ug)

*Adjusted for Substrate Blank

**Using the extinction coefficient $13260 M^{-1}cm^{-1}$

***Using the path correction 0.320 cm

[IDENTIFICATION]

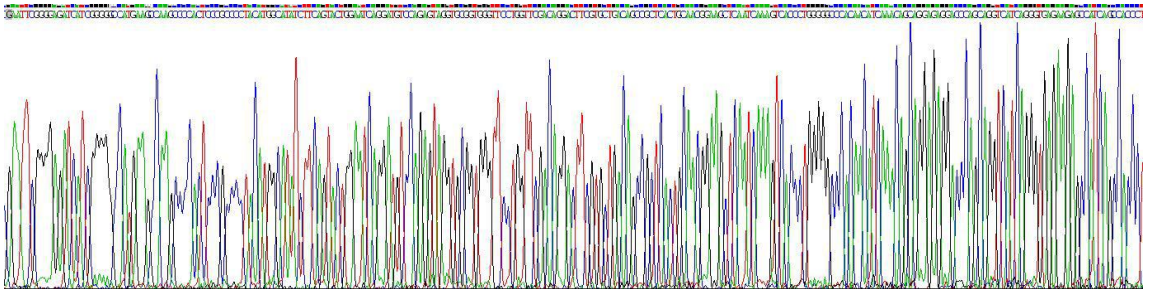


Figure 1. Gene Sequencing (extract)

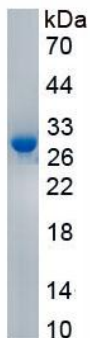


Figure 2. SDS-PAGE

Sample: Active recombinant GZMB, Pig

[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.