

DESCRIPTION

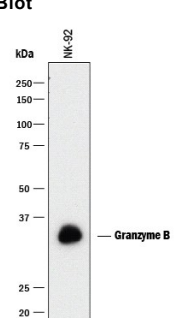
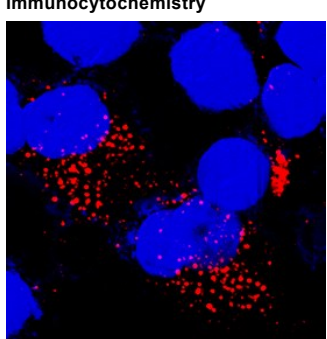
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|---------------------------|---|
| Species Reactivity | Human |
| Specificity | Detects human Granzyme B in direct ELISAs and Western blots. Does not cross-react with recombinant human (rh) Granzyme A, rhGranzyme H, recombinant mouse (rm) Granzyme B, rmGranzyme C, rmGranzyme D, or rmGranzyme G. |
| Source | Monoclonal Mouse IgG _{2A} Clone # 351927 |
| Purification | Protein A or G purified from hybridoma culture supernatant |
| Immunogen | Mouse myeloma cell line NS0-derived recombinant human Granzyme B Gly19-Tyr247 Accession # P10144 |
| Formulation | Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied as a 0.2 µm filtered solution in PBS. |

APPLICATIONS

Please Note: Optimal dilutions should be determined by each laboratory for each application. *General Protocols* are available in the *Technical Information* section on our website.

| | Recommended Concentration | Sample |
|---|--|--|
| Western Blot | 0.5 µg/mL | See Below |
| Immunocytochemistry | 8-25 µg/mL | See Below |
| Intracellular Staining by Flow Cytometry | 2.5 µg/10 ⁶ cells | NK-92 human natural killer lymphoma cell line fixed with paraformaldehyde and permeabilized with saponin |
| CyTOF-ready | Ready to be labeled using established conjugation methods. No BSA or other carrier proteins that could interfere with conjugation. | |

DATA

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| <p>Western Blot</p>  <p>Detection of Human Granzyme B by Western Blot. Western blot shows lysate of NK-92 human natural killer lymphoma cell line. PVDF membrane was probed with 0.5 µg/mL of Mouse Anti-Human Granzyme B Monoclonal Antibody (Catalog # MAB2906) followed by HRP-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # HAF018). A specific band was detected for Granzyme B at approximately 32-35 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.</p> | <p>Immunocytochemistry</p>  <p>Granzyme B in Human PBMCs. Granzyme B was detected in immersion fixed human peripheral blood mononuclear cells (PBMCs) using Mouse Anti-Human Granzyme B Monoclonal Antibody (Catalog # MAB2906) at 8 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Mouse IgG Secondary Antibody (red; Catalog # NL007) and counterstained with DAPI (blue). Specific staining was localized to cytoplasm. View our protocol for Fluorescent ICC Staining of Non-adherent Cells.</p> |
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PREPARATION AND STORAGE

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| Reconstitution | Reconstitute at 0.5 mg/mL in sterile PBS. |
| Shipping | The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C |
| Stability & Storage | Use a manual defrost freezer and avoid repeated freeze-thaw cycles. <ul style="list-style-type: none"> • 12 months from date of receipt, -20 to -70 °C as supplied. • 1 month, 2 to 8 °C under sterile conditions after reconstitution. • 6 months, -20 to -70 °C under sterile conditions after reconstitution. |

BACKGROUND

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 (1, 2). Granzyme B plays an essential role in granule-mediated apoptosis and may have additional roles in rheumatoid arthritis and in bacterial and viral infections (3). It activates various caspases and cleaves proteins such as aggrecan (3). Human Granzyme B is synthesized as a precursor (247 residues) with a signal peptide (residues 1-18), a pro peptide (residues 19-20), and a mature chain (residues 21-247) (4-6). The recombinant human (rh) Granzyme B consisting of residues 19-247 was expressed and purified. After being activated by active cathepsin C, rhGranzyme B cleaves a thioester substrate described previously (3).

References:

1. Kam, C-M. *et al.* (2000) *Biochim. Biophys. Acta* **1477**:307.
2. Smyth, M.J. *et al.* (1996) *J. Leukoc. Biol.* **60**:555.
3. Froelich, C.J. (2004) in *Handbook of Proteolytic Enzymes*, Barrett, A.J. *et al.* eds. pp. 1549.
4. Schmid, J. and C. Weissman (1987) *J. Immunol.* **139**:250.
5. Caputo, A. *et al.* (1988) *J. Biol. Chem.* **263**:6363.
6. Trapani, J.A. *et al.* (1988) *Proc. Natl. Acad. Sci. USA* **85**:6924.