

IOTest Anti-Granzyme B-FITC

PN B46038 – 1 mL – Liquid – Clone GB11

Analyte Specific Reagent.

Analytical and performance characteristics are not established

IOTest

Anti-Granzyme B-FITC

PN B46038

Conjugated Antibody
Liquid - 1 mL

Specifications	
Clone	GB11
Hybridoma	SP2/0 x balb/c
Immunogen	Human Granzyme B, derived from NK cell line YT-INDY
Isotype	IgG1
Species	Mouse
Source	Purified
Purification	Affinity chromatography
Fluorochrome	Fluorescein isothiocyanate (FITC)
Molar ratio	FITC / Ig: 2.9 - 5.5
λ excitation	488 nm
Emission Peak	525 nm
Buffer	PBS pH 7.2 plus 2 mg / mL BSA and 0.1% NaN ₃

SPECIFICITY

Granzyme B (GrB) is single chain and single domain serine protease. GrB is member of the chymotrypsin superfamily. GrB is synthesised as an inactive preproenzyme and transported into the endoplasmatic reticulum (ER) as proGrB. ProGrB, covalently modified with a mannose-6-phosphate (M6P) group, is transported in ER-derived vesicles to the Golgi apparatus (GA). Within the secretory granules, granzymes are stored in association with the chondroitin sulphate containing proteoglycan serglycin (SG). The GrB molecule alone has a high positive surface charge, but when GrB binds to SG its charge may be substantially neutralized (1, 2, 3, 4).

The newly synthesised GrB is heterogeneously glycosylated. The mature enzyme has two potential glycosylation sites. The process of GrB glycosylation results in generation of both the 32 and 35 kDa glycosylated forms of GrB. The 32 kDa GrB forms contain high mannose oligosaccharide moieties and accumulate in cytotoxic T lymphocytes (CTLs) after T cell receptor (TCR) stimulation. In contrast, the 35 kDa GrB forms, which possess only the complex oligosaccharide groups, are not stored in CTLs and instead they are secreted through the constitutive calcium-independent secretory pathway after TCR activation (4, 5, 6).

GrB is the most abundant serine protease stored in secretory granules of CTLs and NK cells. GrB can be produced by plasmacytoid dendritic cells (pDCs) (7).

GrB-induced cell death is a primary mechanism in cytotoxic T lymphocytes (CTLs) and natural killer (NK) cells to eliminate harmful target cells including allogeneic, virally infected and tumor cells.

This mechanism implies activation of several pro-apoptotic pathways by direct proteolysis. The mannose 6-phosphate receptor has been identified as the plasma membrane receptor for GrB (6, 8).

The monoclonal antibody (mAb) GB11 recognizes the human, mouse and rat GranzymeB (9, 10, 11).

REAGENT CONTENTS

Concentration: See lot specific Certificate of Analysis at www.beckmancoulter.com.

PRECAUTIONS

1. This reagent contains 0.1% sodium azide. Sodium azide under acid conditions yields hydrazoic acid, an extremely toxic compound. Azide compounds should be flushed with running water while being discarded. These precautions are recommended to avoid deposits in metal piping in which explosive conditions can develop. If skin or eye contact occurs, wash excessively with water.
2. Specimens, samples and all material coming in contact with them should be considered potentially infectious and disposed of with proper precautions.
3. Never pipet by mouth and avoid contact of samples with skin and mucous membranes.
4. Do not use antibody beyond the expiration date on the label.
5. Do not expose reagents to strong light during storage or incubation.
6. Avoid microbial contamination of reagents or incorrect results might occur.
7. Use good laboratory practices when handling this reagent.
8. Any change in the physical appearance of the reagents may indicate deterioration and the reagent should not be used.

STORAGE AND HANDLING CONDITIONS AND STABILITY

This reagent is stable up to the expiration date when stored at 2 – 8°C. Do not freeze. No reconstitution is necessary. This monoclonal antibody may be used directly from the vial. Bring reagent to 18 – 25°C prior to use.

SELECTED RESEARCH REFERENCES

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4. Shi L, Keefe D, Durand E, Feng H, Zhang D and Lieberman J: Granzyme B binds to target cells mostly by charge and must be added at the same time as perforin to trigger apoptosis. *J Immunol* 174: 5456-5461, 2005.
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7. Jahrsdorfer B. et al, Granzyme B produced by human plasmacytoid dendritic cells suppresses T-cell expansion, *Blood*, 2010, 11, 115, 6.
8. Grifliths GM and Isaaz S., Granzymes A and B Are Targeted to the Lytic Granules of Lymphocytes by the Mannose-6-Phosphate Receptor, *The Journal of Cell Biology*, 1993, 120, 4, 885-896.
9. Thiery et al., Perforin activates clathrin- and dynamin-dependent endocytosis, which is required for plasma membrane repair and delivery of granzyme B for granzyme-mediated apoptosis, *Blood*, 2010, 115(8), 1582–1593.
10. Wagner et al., Expression of granzyme B in peripheral blood polymorphonuclear neutrophils (PMN), myeloid cell lines and in PMN derived from haematopoietic stem cells in vitro, *Molecular Immunology* 2008, 45, 1761–1766.

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