

Anti- Granzyme B

Catalog No.	Description
AMB35-5M	6 ml of Ready-to-Use Antibody for use with BioGenex Super Sensitive™ Detection Systems OR equivalent detection system
AMB35-10M	10 ml of Ready-to-Use Antibody in a barcode labeled vial for use with BioGenex Super Sensitive™ Detection Systems and i6000™ Automated Staining Systems
MUB35-UC	1 ml of Concentrated Antibody for use with BioGenex Super Sensitive™ Detection Systems OR equivalent detection system
MUB35-5UC	0.5 ml of Concentrated Antibody for use with BioGenex Super Sensitive™ Detection Systems OR equivalent detection system
AXB35-YCD	Ready-to-Use Antibody in Barcode labeled vial for use on the Xmatrx® Elite/Ultra Staining System, 160 tests
AXB35-50D	Ready-to-Use Antibody in Barcode labeled vial for use on the Xmatrx® Elite/Ultra Staining System, 50 tests

Clone	Species	Ig Class
2C5	Mouse	IgG2a

Intended Use

For In Vitro Diagnostic Use. This antibody is designed for the specific localization of Granzyme B in formalin-fixed, paraffin-embedded (FFPE) tissue sections. Evaluation must be performed by a qualified pathologist.

Summary and Explanation

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. They are synthesized, processed and released by exocytosis in lysosome-like granules containing perforin. Granzyme B has the strongest apoptotic activity of all the granzymes as a result of its caspase-like ability to cleave substrates at aspartic acid residues thereby activating procaspases directly and cleaving downstream caspase substrates. Recent studies have demonstrated that the expression of granzyme B by Immunohistochemistry in several entities of extranodal peripheral T cell lymphoma (PTCL) and NK cell lymphomas, including nasal and nasal-type NK/T cell lymphomas, hepatosplenic and non-hepatosplenic PTCL, enteropathy-type (ETCL) and non-ETCL intestinal PTCL, subcutaneous panniculitis-like PTCL (SPTCL), cutaneous CD8+ epidermotropic lymphomas as well as in nodal and cutaneous CD30+ anaplastic large cell lymphomas (ALCL). In contrast, only a few nodal PTCL-unspecified (UC) express cytotoxic phenotype while angio immunoblastic (AILD) lymphomas do not. The expression of granzyme B may be important for the identification and classification of extranodal T- and NK-cell

lymphomas since many of these tumors do not have specific morphology and phenotype.

Storage and Handling

Store at 2-8°C. Fresh dilutions, if required, should be prepared prior to use and are stable and steady for up to one day at room temperature (20-26°C). Diluted antibody preparations can be refrigerated or frozen for extended shelf life.

Principles of the Procedure

Antigen detection by immunohistochemistry (**IHC**) is a two-step process wherein the primary antibody binds to the antigen of interest and that binding is detected by a chromogen. The **primary antibody** may be used in IHC using manual techniques or BioGenex Automated Staining System. Positive and negative controls should always be run simultaneously with all patient specimens.

Reagents Provided

Mouse Monoclonal Antibody Granzyme B is affinity purified and diluted in PBS, pH 7.2, containing 1% BSA and 0.09% sodium azide.

Dilution of Primary Antibody

BioGenex Ready-to-Use antibodies have been optimized for use with the recommended BioGenex Detection System and should not require further dilution.

BioGenex concentrated antibodies must be diluted in accordance with the recommended protocol when used with the recommended BioGenex Detection System.

Recommended Protocol

Refer to the following table for conditions specifically recommended for this antibody. Refer to the BioGenex website for guidance on specific staining protocols or other requirements.

Parameter	BioGenex Recommendations
Control Tissue	Spleen tissue as available with Biogenex FB-B35M* & FG-B35M*
Recommended Dilution for Concentrated Antibody	1:10-25 in HK941
Recommended Pretreatment (Manual/i6000)**	EZ-AR2 (HK522-XAK)
Recommended Pretreatment (Xmatrx)	EZ-AR2 Elegance (HX032-YCD)
Antibody Incubation (Manual/i6000)	30-60 Min at RT
Antibody Incubation (Xmatrx)	30-60 Min at 25°C

Detection System for Manual, Xmatrx & i6000 systems***	Use BioGenex Two-Step OR One-Step Super Sensitive™ Polymer-HRP IHC Detection System/DAB; see p. 2 for more information
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*FB: positive control micro chamber slides, FG: positive control microscopic slides. Xmatrx requires micro chamber slides.

**Pretreatment times will vary based on individual microwave power.

***For automation systems (Xmatrx-Elite, Xmatrx-Ultra & i6000 Diagnostics), refer to the factory protocols provided with the instrument.

Detection System	Two-Step HRP Kit	One-Step HRP Kit	Link and Label Kit
Manual	QD440-XAKE (1000 Test)	QD630-XAKE (1000 Test)	QP300-XAKE (1000 Test)
	QD430-XAKE (1000 Test)		
	QD420-YIKE (500 Test)	QD620-XAKE (500 Test)	QP900-9LE (500 Test)
	QD400-60KE (60 Test)		
Xmatrx - Automation	QD550-YCDE (200 Test)	QD610-YADE (200 Test)	N/A
i6000 - Automation	QD410-YAXE (200 Test)	QD610-YAXE (200 Test)	N/A
For more information, visit www.biogenex.com .			

Precautions

This product contains sodium azide at concentrations of less than 0.1%. Sodium azide is not classified as a hazardous chemical at the product concentrations, but proper handling protocols should be observed. For more information, a Safety Data Sheet (SDS) for sodium azide is available upon request. Dispose of unused reagents according to Local, State and Federal Regulations. Wear suitable Personal Protective Equipment, do not pipette reagents by mouth, and avoid contact of reagents and specimens with skin and mucous membranes. If reagents or specimens come in contact with sensitive area, wash with copious amounts of water.

Quality Control

Refer to BioGenex detection system documents for guidance on general quality control procedures.

Troubleshooting

Refer to the troubleshooting section in the documentation for BioGenex Detection Systems (or equivalent detection systems) for remedial actions on detection system related issues, or contact BioGenex Technical Support Department at 1-800-421-4149 or support@biogenex.com or your local distributor to report unusual staining.

Expected Results

This antibody stains membrane and cytoplasm in positive cells in formalin-fixed, paraffin embedded tissue sections. An example image of a tissue section stained with this antibody can be found on the product page on the BioGenex website. Interpretation of the staining result is solely the responsibility of the user. Experimental results should be confirmed by a medically-established diagnostic product or procedure.

Limitations of the Procedure

Improper tissue handling and processing prior to immunostaining can lead to inconsistent results. Variations in embedding and fixation or the nature of the tissue may lead to variations in results. Endogenous peroxidase activity or pseudo peroxidase activity in erythrocytes and tissue biotin may result in non-specific staining based on the detection system employed. Tissues containing Hepatitis B Surface Antigen (HBsAg) may give false positive with horseradish peroxidase systems. Improper counterstaining and mounting may compromise the interpretation of results.

Bibliography

1. Trapani, J.A. (2001) *Genome Biol* 2, REVIEWS3014.
2. Lord, S.J. et al. (2003) *Immunol. Rev.* 193, 31-8.
3. Trapani, J.A. and Sutton, V.R. (2003) *Curr. Opin. Immunol.* 15, 533-43.

	Temperature Limitation		In Vitro Diagnostic Medical Device
	Use By Date		Batch Code
	Non-Sterile		Consult Instructions for Use
	Representative in the European Community		Manufacturer

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Category	Antibodies	Revision No.	D
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