

Anti-ZAP-70 [ZAP70-C3]

Catalog No.	Description	
AM544-5M	6 ml of Ready-to-Use Antibody for use with BioGenex Super Sensitive TM Detection Systems OR equivalent detection system	
AM544-10M	M 10 ml of Ready-to-Use Antibody in a barcode labeled vial for use with BioGenex Super Sensitive TM Detection Systems and i6000 TM Automated Staining Systems	
MU544-UC	1 ml of Concentrated Antibody for use with BioGenex Super Sensitive TM Detection Systems OR equivalent detection system	
MU544-5UC	0.5 ml of Concentrated Antibody for use with BioGenex Super Sensitive TM Detection Systems OR equivalent detection system	
AX544-YCD	Ready-to-Use Antibody in Barcode labeled	
AX544-50D	Ready-to-Use Antibody in Barcode labeled vial for use on the Xmatrx [®] Elite/Ultra Staining System, 50 tests	

Clone	Species	Ig Class
ZAP70-C3	Mouse	IgG2a

Intended Use

For In Vitro Diagnostic Use. This antibody is designed for the specific localization of ZAP-70 in formalin-fixed, paraffinembedded (FFPE) tissue sections. Evaluation must be performed by a qualified pathologist.

Summary and Explanation

ZAP-70 is an abbrevation for Zeta-chain-associated protein kinase 70 (70 is the molecularweight in kDa). The protein is a member in the protein-tyrosine kinase family. ZAP-70 isnormally expressed in T cells and natural killer cells and has a critical role in the initiation of T-cell signaling. ZAP-70 in B cells is used as a prognostic marker in identifying different forms of chronic lymphocytic leukemia (CLL). ZAP-70 protein is expressed in leukemic cells of approximately 25% of Chronic Lymphocytic Leukemia (CLL) cases as well. ZAP-70 expression is an excellent surrogate marker for the distinction between the Ig-mutated (ZAP-70 negative) and Ig-unmutated (ZAP-70 positive).

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.

Emergo Europe, Prinsessegracht 20, 2514AP The Hague, The Netherlands

Principles of the Procedure

Antigen detection by immunohistochemistry (<u>IHC</u>) is a two-step process wherein the primary antibody binds to the antigen of interest and that binding is detected by a chromogen. The <u>primary</u> <u>antibody</u> 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 to ZAP70is 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	TONSILas available from BioGenex FB-544M* & FG- 544M*	
Recommended Dilution for Concentrated Antibody	1:50-100 in HK941	
Recommended Pretreatment (Manual/i6000)**	EZ-AR2 (HK522-XAK)	
Recommended Pretreatment (Xmatrx)	EZ-AR2 Elegance (HX032- YCD)	
Antibody Incubation (Manual/i6000)	30-60 mins at RT	
Antibody Incubation (Xmatrx)	30-60 mins at RT	
Detection System for Manual, Xmatrx & i6000 systems***	Use BioGenex Two-Step OR One-Step Super Sensitive TM Polymer-HRP IHC Detection System/DAB; see p. 2 for more information	

*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.

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

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

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

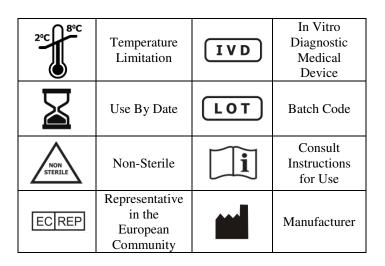
This antibody stains cytoplasm in positive cells in formalinfixed, paraffin embedded tissue sections. An example image of a tissue section stained with this antibody can be found on the product page on the BioGenex website. Interpretation of the staining result is solely the responsibility of the user. Experimental results should be confirmed by a medicallyestablished 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

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- Center for Disease Control. Decontamination of Laboratory Sink Drains to Remove Azide Salts. Center for Disease Control Manual Guide--Safety Management,No. CDC-22, Atlanta, Georgia. April 30, 1976.
- 5. Kiernan JA. Histological and Histochemical Methods: Theory and Practice. New York: Pergamon Press 1981.
- 6. Nadji M, Morales AR. Immunoperoxidase, part 1: the techniques and its pitfall. Lab Med 1983; 14:767-770.



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