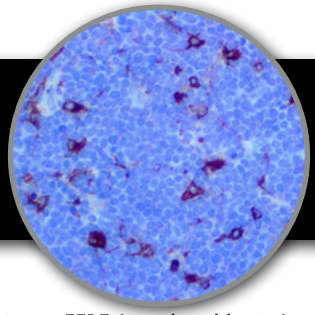
MMP-9, RMab

Clone: EP127

Rabbit Monoclonal

CE IVD



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Inset: IHC of MMP-9 on a FFPE Lymphomblastic Lymphoma Tissue

Intended Use

For In Vitro Diagnostic Use.

This antibody is intended for use in Immunohistochemical applications on formalin-fixed paraffin-embedded tissues (FFPE), frozen tissue sections and cell preparations. Interpretation of results should be performed by a qualified medical professional.

* The MMP-9 antibody, clone EP127, has been manufactured using Epitomics RabMab® technology covered under Patent No.'s 5,675,063 and 7,402,409.

Immunogen

A synthetic peptide corresponding to residues of human MMP-9 protein.

Summary and Explanation

The matrix metalloproteinases (MMPs) are responsible for degradation of the extracellular matrix. The MMPs and their specific tissue inhibitor metalloproteinases (TIMP) have been associated with tumor cell invasion and metastasis in a number of adult tumors. MMP-9, also designated as 92-kDa Type IV Collagenase or gelatinase B, is a member of MMPs, which is produced as a 92- kDa pro-enzyme by neutrophils, macrophages, mast cells and stromal cells, as a normal constituent and released into the extracellular environment after activation in inflammatory tissues.

MMP-9 may be involved in the development of several human malignancies, as degradation of collagen IV in basement membrane and extracellular matrix facilitates tumor progression, including invasion, metastasis, growth and angiogenesis. The expression levels of MMP-9 in tumors are elevated compared with the corresponding normal tissues in a variety of cancer types, including breast, colon, gastric and nasopharyngeal cancers. MMP-9 may play an important role in angiogenesis and neovascularization. For example, MMP9 appears to be involved in the remodeling associated with malignant glioma neovascularization. Increased expression has been seen in a metastatic mammary cancer cell line.

Antibody Type	Rabbit Monoclonal	**Clone**	EP127
Isotype	IgG	**Reactivity**	Paraffin, Frozen
Localization	Cytoplasmic	**Control**	Tonsil, Spleen, Lymphoblastic Lymphoma
Species Reactivity		Human, Predicted: Rat	

Presentation

MMP-9 is a rabbit monoclonal antibody derived from cell culture supernatant that is concentrated, dialyzed, filter sterilized and diluted in buffer pH 7.5, containing BSA and sodium azide as a preservative.

Presentations

Catalog Num.	Antibody Type	Dilution	Volume/Qty
BSB 2538	Tinto Prediluted	Ready-to-Use	3.0 mL
BSB 2539	Tinto Prediluted	Ready-to-Use	7.0 mL
BSB 2540	Tinto Prediluted	Ready-to-Use	15.0 mL
BSB 2541	Concentrated	1:50 - 1:200	0.1 mL
BSB 2542	Concentrated	1:50 - 1:200	0.5 mL
BSB 2543	Concentrated	1:50 - 1:200	1.0 mL
BSB 2544	Control Slides	Not Applicable	5 slides

Precautions

1. For professional users only. Ensure results are interpreted by a medical professional.
2. This product contains sodium azide (NaN3), a toxic chemical which may react with plumbing to form highly explosive build-ups of metal azides. Upon disposal, flush with large volumes of water to prevent sodium azide build-up.
3. Ensure proper handling procedures are used with reagent. Always wear proper laboratory equipment such as laboratory coat and gloves when handling reagents.
4. Unused solution should be disposed of according to local and federal regulations.
5. Do not ingest reagent. If reagent ingested, contact a poison control center immediately.
6. For complete recommendations for handling biological specimens please refer to the CDC document, "Guidelines for Safe Work Practices in Human and Animal Medical Diagnostic Laboratories" (8).

Storage

Store at 2-8 °C. Do not use after expiration date listed on package label. Temperature fluctuations should be avoided. Store appropriately when not in use, and avoid prolonged exposure to room temperature conditions.

Specimen Preparation

Paraffin sections: The antibody can be used on formalin-fixed paraffin-embedded (FFPE) tissue sections. Ensure tissue undergoes appropriate fixation to ensure best results. Pre-treatment of tissues with heat-induced epitope retrieval (HIER) is recommended using Bio SB ImmunoDNA Retriever with Citrate (BSB 0020-BSB 0023), ImmunoDNA Retriever with EDTA (BSB 0030-BSB 0033) or ImmunoDNA Digestor (BSB 0108-0112). See reverse side for complete protocol. Tissue should remain hydrated via use of Bio SB Immuno/DNA Washer solutions (BSB 0029 & BSB 0042).
Frozen sections and cell preparations: The antibody can be used for labeling acetone-fixed frozen sections and acetone-fixed cell preparations.

Staining Procedure

1. Cut and mount 3-5 micron formalin-fixed paraffin-embedded tissues on positive charged slides such as Bio SB Hydrophilic Plus Slides (BSB 7028).
2. Air dry for 2 hours at 58° C.
3. Deparaffinize, dehydrate and rehydrate tissues.
4. Subject tissues to heat epitope retrieval using a suitable retrieval solution such as ImmunoDNA Retriever with Citrate (BSB 0020-BSB 0023) or EDTA (BSB 0030-BSB 0033).
5. Any of three heating methods may be used:

a. TintoRetriever Pressure Cooker or Equivalent

Place tissues/slides in a staining dish or coplin jar containing the ImmunoDNA Retriever with Citrate or EDTA, and place in the pressure cooker. Add 1-2 inches of distilled water to the pressure cooker and turn heat to high. Incubate for 15 minutes. Open and immediately transfer slides to room temperature.

b. TintoRetriever PT Module or Water Bath Method

Place tissues/slides in a pre-warmed staining dish or coplin jar containing the ImmunoDNA Retriever with Citrate or EDTA at 95°-99° C. Incubate for 30-60 minutes.

c. Conventional Steamer Method

Place tissues/slides in a pre-warmed staining dish or coplin jar containing the ImmunoDNA Retriever with Citrate or EDTA in a Steamer, cover and steam for 30-60 minutes.

6. After heat treatment, transfer slides in ImmunoDNA Retriever with Citrate or EDTA to room temperature and let stand for 15-20 minutes.
7. For manual staining, perform antibody incubation at ambient temperature. For automated staining methods, perform antibody incubation according to instrument manufacturer's instructions.
8. Wash slides with IHC wash buffer or DI water.
9. Continue IHC staining protocol.

Recommended IHC Protocol

Step	ImmunoDetector AP/HRP	PolyDetector AP/HRP	PolyDetector Plus HRP
Peroxidase/AP Blocker	5 min.	5 min.	5 min
Primary Antibody	30-60 min.	30-60 min.	30-60 min.
1st Step Detection	10 min.	30-45 min.	15 min.
2nd Step Detection	10 min.	Not Applicable	15 min.
Substrate-Chromogen	5-10 min.	5-10 min.	5-10 min.
Counterstain	Varies	Varies	Varies

Performance Characteristics

Normal Tissues	
Positive (+)	
Tonsil	5/5 (100%)
Spleen	5/5 (100%)
Abnormal Tissues	
Positive (+)	
Lymphoblastic Lymphoma	6/6 (100%)
NH Lymphoma	3/3 (100%)
Breast Carcinoma	1/3 (33%)

Product Limitations

Due to inherent variability present in immunohistochemical procedures (including fixation time of tissues, dilution factor of antibody, retrieval method utilized and incubation time), optimal performance should be established through the use of positive and negative controls. Results should be interpreted by a medical professional.

References

1. Vandooren, J; Van den Steen, PE; Opdenakker, G. "Biochemistry and molecular biology of gelatinase B or matrix metalloproteinase-9 (MMP-9) The next decade". Crit Rev Biochem Mol Biol. 2013; 48 (3): 222–72.
2. Forsyth PA, et al. "Gelatinase-A (MMP-2), gelatinase-B (MMP-9) and membrane type matrix metalloproteinase-1 (MT1-MMP) are involved in different aspects of the pathophysiology of malignant gliomas". British Journal of Cancer. 1999; 79 (11-12).
3. Heissig B, et al. "Recruitment of Stem and Progenitor Cells from the Bone Marrow Niche Requires MMP-9 Mediated Release of Kit-Ligand". Cell. 2002;109 (5): 625–37.
4. Morini M, et al. "The α3β1 integrin is associated with mammary carcinoma cell metastasis, invasion, and gelatinase B (mmp-9) activity". International Journal of Cancer, 2000; 87 (3): 336–42.
5. Farina AR, Mackay AR. "Gelatinase B/MMP-9 in Tumour Pathogenesis and Progression". Cancers (Basel), 2014; 6 (1): 240–96.
6. Zucker S, et al. "Plasma assay of gelatinase B: tissue inhibitor of metalloproteinase complexes in cancer". Cancer , 1995; 76 (4): 700–708.
7. Groblewska M, Set al. "The role of matrix metalloproteinases (MMPs) and their inhibitors (TIMPs) in the development of esophageal cancer". Folia Histochem. Cytobiol. 2012; 50 (1): 12–9.
8. U.S. Department of Health and Human Services: Centers for Disease Control and Prevention. Guidelines for Safe Work Practices in Human and Animal Medical Diagnostic Laboratories. Supplement / Vol. 61, January 6, 2012.

Symbol Key / Légende des symboles/Erläuterung der Symbole

EC REP EMERGO EUROPE Prinsessegracht 20 2514 AP The Hague The Netherlands	8°C / 2°C Storage Temperature Limites de température Zulässiger Temperaturbereich	Manufacturer Fabricant Hersteller	REF Catalog Number Référence du catalogue Bestellnummer
IVD In Vitro Diagnostic Medical Device Dispositif médical de diagnostic in vitro In-Vitro-Diagnostikum	Read Instructions for Use Consulter les instructions d'utilisation Gebrauchsanweisung beachten	Expiration Date Utiliser jusque Verwendbar bis	LOT Lot Number Code du lot Chargenbezeichnung

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