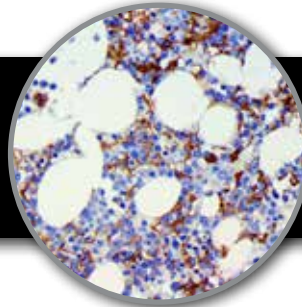


CD16, RMab

Clone: EP364

Rabbit Monoclonal



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Inset: IHC of CD16 on a FFPE Bone Marrow 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 CD16 antibody, clone EP364, has been manufactured using Epitomics RabMab® technology covered under Patent No's 5,675,063 and 7,402,409.

Immunogen

Synthetic peptide corresponding to residues of human CD16 protein.

Summary and Explanation

CD16 is a low affinity Fc receptor, found on the surface of natural killer cells, neutrophil polymorphonuclear leukocytes, monocytes and macrophages. CD16 has been identified as Fc receptors FcγRIIIa (CD16a) and FcγRIIIb (CD16b). These receptors bind to the Fc portion of IgG antibodies which then activates NK cells for antibody-dependent cell-mediated cytotoxicity. A lack of CD16 in a given population of neutrophils may indicate prematurity, as could be caused by a left shift due to neutrophilic leukocytosis induced by tissue necrosis or bacterial infection.

The IHC of CD16 is useful in the differential diagnosis of hepatosplenic gamma delta T-cell lymphoma and gamma delta T-cell large granular lymphocyte leukemia from other peripheral T-cell lymphomas, such as mucosal and cutaneous gamma delta T-cell lymphoma. A significant decrease can be seen in the number of granulocytes expressing CD16 in chronic myelomonocytic leukemia compared to chronic myelogenous leukemia and control bone marrow biopsy, probably related to dysgranulopoiesis. It has also been demonstrated that colorectal carcinoma patients with high CD16+ cell infiltration is associated with improved overall survival after adjusting for known prognostic factors and this association was independent from CD8+ lymphocyte infiltration and presence of metastases.

Antibody Type	Rabbit Monoclonal	Clone	EP364
Isotype	IgG	Reactivity	Paraffin, Frozen
Localization	Cytoplasmic, Membranous	Control	Placenta, Liver, Breast Spleen, Thymus, Lung
Species Reactivity	Human		

Presentation

CD16 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 3321	Tinto Prediluted	Ready-to-Use	3.0 mL
BSB 3322	Tinto Prediluted	Ready-to-Use	7.0 mL
BSB 3323	Tinto Prediluted	Ready-to-Use	15.0 mL
BSB 3324	Concentrated	1:50 - 1:200	0.1 mL
BSB 3325	Concentrated	1:50 - 1:200	0.5 mL
BSB 3326	Concentrated	1:50 - 1:200	1.0 mL
BSB 3327	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" (5).

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





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. Janeway, Charles. Appendix II. CD antigens. Immunobiology (5 ed.) 2001; New York: Garland. ISBN 0-8153-3642-X.
2. Vidranski, V; Laskaj, R; Sikiric, D; Skerk, V. Platelet satellitism in infectious disease?. Biochem Med (Zagreb) 2015;. 25: 285-94.
3. Gibson SE, et al. Natural killer cell subsets and natural killer-like T-cell populations in benign and neoplastic B-cell proliferations vary based on clinicopathologic features. Hum Pathol. 2011; May;42(5):679-87
4. Qubaja M, et al. The detection of CD14 and CD16 in paraffin-embedded bone marrow biopsies is useful for the diagnosis of chronic myelomonocytic leukemia. Virchows Arch. 2009; Apr;454(4):411-9.
5. 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		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 jusqu'à Verwendbar bis	LOT	Lot Number Code du lot Chargenbezeichnung

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