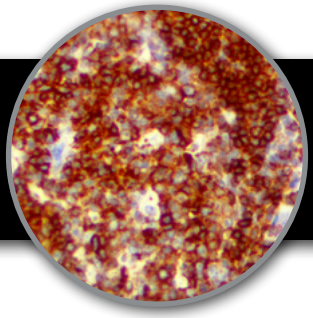
CD45RA

Clone: 4KB5

Mouse Monoclonal

CE IVD



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Inset: IHC of CD45RA on a FFPE Tonsil 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.

Immunogen

Hairy cell leukemia cells.

Summary and Explanation

CD45 is a complex molecule and is comprised of different glycoproteins ranging from 180-240 kDa. Expression of CD45 is found on all hemopoietic cells. Detection of the different isoforms can distinguish between different cell forms (e.g., naive T-cells and memory T-cells). CD45RA is an isoform of the CD45 complex and has restricted expression between different subtypes of lymphoid cells.

CD45RA antibody reacts with mature, non-activated T and B-cells. CD45RA is also reactive with medullary thymocytes, mantle-zone lymphocytes in follicles of lymph nodes, spleen and lymphocytes of the paracortex. CD45RA shows no reactivity with cortical thymocytes, immature T-cells or activated B-cells in germinal centers.

Antibody Type	Mouse Monoclonal	**Clone**	4kB5
Isotype	IgG1/K	**Reactivity**	Paraffin, Frozen
Localization	Membranous	**Control**	Tonsil, Lymph Node
Species Reactivity		Human	

Presentation

CD45RA is a mouse monoclonal antibody derived from cell culture supernatant that is concentrated, dialyzed, fillter 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 5253	Tinto Prediluted	Ready-to-Use	3.0 mL
BSB 5254	Tinto Prediluted	Ready-to-Use	7.0 mL
BSB 5255	Tinto Prediluted	Ready-to-Use	15.0 mL
BSB 5256	Concentrated	1:25 - 1:100	0.1 mL
BSB 5257	Concentrated	1:25 - 1:100	0.5 mL
BSB 5258	Concentrated	1:25 - 1:100	1.0 mL
BSB 5259	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" (3).

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.

Performance Characteristics

Normal Tissues

Positive (+)
formalin-fixed B-cell areas: lymph node, spleen, ileum, colon, tonsil, thymus
lymphocytes of the mantle zone (strongly labelled)
Germinal centre cells (moderate to weakly labelled)
Occasional lymphocytes in the: Dermis, interstitium of the lung, sinusoids of the liver, periportal areas of the liver
Negative (-)
Plasma Cells
frozen tissue or cell suspensions: peripheral blood B cells, subpopulation of T cells, monocytes

Abnormal Tissues

Positive (+)	
B cell lymphomas 30/34	low-grade B-cell lymphomas 15/15
B-cell lymphomas 13/32	T-cell lymphoblastic lymphomas 2/19
T-cell lymphomas: T-zone 3/8 Malignant histiocytosis of the intestine (MHI) 1/5	
T-cell-rich B-cell lymphoma: neoplastic pop (all or a subset)	
lymphocyte-predominant Hodgkin's lymphoma: lymphocytic and histiocytic cells (consistent)	
Negative (-)	
centroblastic lymphomas with sclerosis 2/2	
plasma cell lymphomas 3/4	
T-cell lymphomas: Lymphocytic, Cutaneous, Immunoblastic, Large cell pleomorphic, lymphoblastic	
lymphocyte-predominant Hodgkin's lymphoma: lymphocytic and histiocytic cells (none or few)	
Neoplastic cells from adenocarcinoma Lung, Breast, Colon, Macrophage malignancies, Monocyte malignancies	

References

1. Myskow MW, Krajewski AS, Salter DM, Dobson CM, Miller EP. Paraffin section immunophenotyping of non-Hodgkin's lymphoma, using a panel of monoclonal antibodies. Am J Clin Pathol 1988;90:564-74.
2. Sewell WA, Cooley MA, Hegen M. NL6. CD45 workshop panel report. In: Kishimoto T, Kikutani H, von dem Borne AEG, Goyert SM, Mason DY, Miyasaka M, et al., editors. Leucocyte typing VI. White cell differentiation antigens. Proceedings of the 6th International Workshop and Conference; 1996 Nov 10-14; Kobe, Japan. New York, London: Garland Publishing Inc.; 1997. p. 499-502.
3. 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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