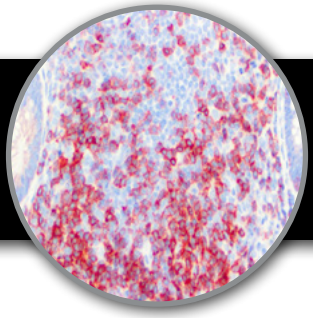
CD45RO

Clone: UCHL-1

Mouse Monoclonal

CE IVD



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Inset: IHC of CD45RO 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

Interleukin-2-dependent human T lymphocytes.

Summary and Explanation

The CD45 family consists of multiple members that are all products of a single complex gene. Three isoforms of CD45 exist: on B-lymphocytes, where the protein is called B220 (its molecular mass is 220 kDA); on naive T-lymphocytes, where it is called CD45RA, and on activated and memory T-lymphocytes, where it is called CD45RO. CD45RO is a single-chain, transmembraneous glycoprotein which represents the low molecular weight isoform of the Leukocyte Common Antigen (LCA). It is expressed on most thymocytes, about 45% of peripheral blood T-cells, virtually all T-cells in skin reactive infiltrates, and the majority of T-cell malignancies. It is also found on a subset of B-cells and on exceptional B-cell Lymphomas.

CD45RO (T-Cell, Pan) antibody reacts with thymocytes and activated T-cells, but only on a subpopulation of resting T-cells. This antibody shows no reactivity with B-cells, making it a good marker for T-cell tumors to be phenotyped. In addition, granulocytes and monocytes are also labeled with this antibody. T-Cell, Pan has been designated as CD45RO at The International Leukocyte Typing Workshop.

Antibody Type	Mouse Monoclonal	**Clone**	UCHL-1
Isotype	IgG2a/K	**Reactivity**	Paraffin, Frozen
Localization	Membranous	**Control**	Tonsil, Lymph Node
Species Reactivity		Human, Mouse, Rat, Non-human primate	

Presentation

CD45RO is a mouse 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 5260	Tinto Prediluted	Ready-to-Use	3.0 mL
BSB 5261	Tinto Prediluted	Ready-to-Use	7.0 mL
BSB 5262	Tinto Prediluted	Ready-to-Use	15.0 mL
BSB 5263	Concentrated	1:250 - 1:1000	0.1 mL
BSB 5264	Concentrated	1:250 - 1:1000	0.5 mL
BSB 5265	Concentrated	1:250 - 1:1000	1.0 mL
BSB 5266	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" (4).

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

References

1 Hall PA, et al. J Clin Path. 1987;40:151-156
2. Smith SH, et al. Immunology. 1986;58:63-70
3. Tworek JA, et al. Am J Clin Pathol. 1998;Nov;110(5):582-589
4. 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.

Performance Characteristics

Normal Tissues

Positive (+)	
thymocytes	mature activated T cells
monocytes	granulocytes
lymphocytes	Membrane of mature myeloid cells
Cortical thymocytes 90%	medullary thymocytes 50%
resting T cells within both the CD4 and CD8 subsets	
Macrophage (cytoplasmic) 20%	
glandular epithelia (cytoplasmic)	Diffuse/weak
squamous epithelium (cytoplasmic)	Diffuse/weak
transitional epithelium (cytoplasmic)	Diffuse/weak
Hepatocytes (cytoplasmic)	Diffuse/weak
Syncytiotrophoblasts (cytoplasmic)	Diffuse/weak
smooth muscle (cytoplasmic)	Diffuse/weak
Negative (-)	
normal B cells	NK cells
cortical thymic blasts	

Abnormal Tissues

Positive (+)	
Mycosis fungoides 100%	Peripheral T-Cell Lymphoma 83%
Reed-Sternberg cells (cytoplasm)	True histiocytic lymphoma 2/2
Malignant histiocytosis of the intestine 100%	
Large B-cell lymphomas of centroblastic type (cytoplasm)	Diffuse
Large B-cell lymphomas of immunoblastic type (cytoplasm)	Diffuse
T acute lymphoblastic lymphoma 78%	
Granulocytic sarcoma: Mature myeloid cells 4/4	
Custaneous malignant T-cell lymphoma: Tumor cells	
Inflammatory skin diseases: Reactive T cells	
Negative (-)	
B-cell lymphomas	Reed-Sternberg cells (membrane)
Cells in cutaneous B-cell lymphomas	
Tumor cells of various non-lymphatic skin tumours	

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.

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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