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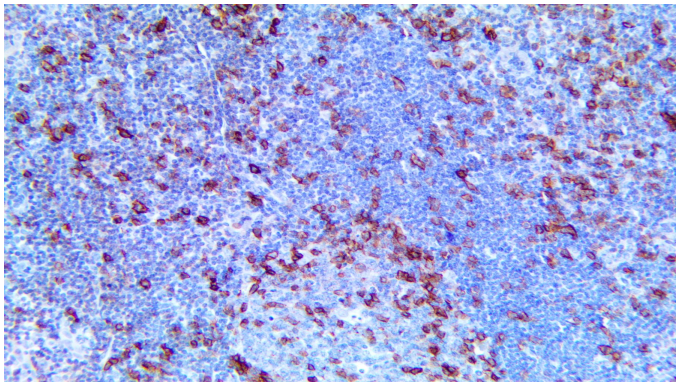
ICOS/CD278

Clone: RM417
Rabbit Monoclonal

CE IVD



Inset: IHC of ICOS/CD278 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
Synthetic peptide corresponding to residues near the C-terminus of the human ICOS protein.

Summary and Explanation
CD278 is also known as inducible T-cell costimulatory molecule (ICOS). CD278 is homologous to the CD28/CTLA-4 proteins and is expressed on activated T cells and unstimulated thymocytes. CD278 plays a major role in regulation of cell-cell signaling, adaptive immune response, and cell proliferation. Interaction of CD278 and its ligand ICOS-L results in increased production of interleukins, which promote differentiation of Tfh and Tregs and development of Th1, Th2, and Th17 cells.

CD278/ICOS-L interaction has been shown to promote either antitumor T cell response or pro-tumor responses when triggered in Tregs (such as in Multiple Myeloma, Acute Myeloid Leukemia, and some invasive Breast Carcinomas). Therefore, both agonistic and antagonistic monoclonal antibodies targeting this pathway can be potential cancer immunotherapy.

ICOS is primarily expressed on activated CD4+ and CD8+ T cells where it regulates immune responses and plays a role in the regulation of T Follicular helper cells. ICOS is a sensitive marker for identifying T cell Lymphomas of Follicular Helper T cell origin, especially certain patterns of Angioimmunoblastic T-cell lymphoma (AITL) and Peripheral T-cell lymphomas with T-follicular Helper Phenotype (PTCL-TFH). This is becoming increasingly important given WHO 2016 diagnostic guidelines. Immunohistochemical analysis revealed that ICOS is widely expressed by malignant cells in skin biopsy specimens from patients with Mycosis Fungoides and Sézary syndrome (SS), as well as involved in node biopsy specimens from patients with SS.

Antibody Type	Rabbit Monoclonal	**Clone**	RM417
Isotype	IgG	**Reactivity**	Paraffin, Frozen
Localization	Membranous	**Species Reactivity**	Human
Control	Colon, Tonsil, Testis, Transitional Cell Carcinoma, T Cell Lymphoblastic Lymphoma		
Application	Immunotherapy, Leukemia and Histiocytic Cancer, Lymphoma, Breast Cancer, Melanoma and Skin Cancer		

Presentation
Anti-ICOS/CD278 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.

Catalog No.	*Presentation*	*Dilution*	*Volume*
BSB-3731-3	Predilute	Ready-to-Use	3.0 mL
BSB-3731-7	Predilute	Ready-to-Use	7.0 mL
BSB-3731-15	Predilute	Ready-to-Use	15.0 mL
BSB-3731-01	Concentrate	1:50-1:200	0.1 mL
BSB-3731-05	Concentrate	1:50-1:200	0.5 mL
BSB-3731-1	Concentrate	1:50-1:200	1.0 mL

Control Slides Available

Catalog No.	*Quantity*
BSB-3731-CS	5 slides

Storage Store at 2-8°C (Control Slides: Store at 20-25°C)

Precautions
1. For professional users only. Results should be interpreted by a qualified medical professional.
2. This product contains <0.1% sodium azide (NaN_3) as a preservative. Ensure proper handling procedures are used with this reagent.
3. Always wear personal protective equipment such as a laboratory coat, goggles, and gloves when handling reagents.
4. Dispose of unused solution with copious amounts of water.
5. Do not ingest reagent. If reagent is ingested, seek medical advice immediately.
6. Avoid contact with eyes. If contact occurs, flush with large quantities of water.
7. Follow safety precautions of the heating device used for epitope retrieval (TintoRetriever Pressure Cooker or similar).
8. For additional safety information refer to Safety Data Sheet for this product.
9. 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" (see References in this document).

Stability
This product is stable up to the expiration date on the product label.
Do not use after expiration date listed on the 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 for 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 on acetone-fixed frozen sections and acetone-fixed cell preparations.

IHC Protocol

1. Cut and mount 3-5 micron formalin-fixed paraffin-embedded tissues on positively 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 induced epitope retrieval (HIER) 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 on trivet 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 IHC, perform antibody incubation at ambient temperature. For automated IHC methods, perform antibody incubation according to instrument manufacturer's instructions.
8. Wash slides with ImmunoDNA washer or DI water.
9. Continue IHC protocol. Wash slides between each step with ImmunoDNA washer solution.

Abbreviated Immunohistochemical 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 / Coverslip	Varies	Varies	Varies

Mounting Protocols

For detailed instructions using biodegradable permanent mounting media such as XyGreen PermaMounter (BSB 0169-0174) or organic solvent based resin such as PermaMounter (BSB 0094-0097), refer to PI0174 or PI0097.

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 qualified medical professional.

References

1. Solinas C, Gu-Trantien C, Willard-Gallo K. The rationale behind targeting the ICOS-ICOS ligand costimulatory pathway in cancer immunotherapy. ESMO Open. 2020;5(1):e000544. doi:10.1136/esmoopen-2019-000544
2. Marinelli O, Nabissi M, Morelli MB, Torquati L, Amantini C, Santoni G. ICOS-L as a Potential Therapeutic Target for Cancer Immunotherapy. Curr Protein Pept Sci. 2018;19(11):1107-1113. doi:10.2174/1389203719666180608093913
3. Wang B, Jiang H, Zhou T, et al. Expression of ICOSL is associated with decreased survival in invasive breast cancer. PeerJ. 2019;7:e6903. Published 2019 May 16. doi:10.7717/peerj.6903
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5. Amatore F, Ortonne N, Lopez M, et al. ICOS is widely expressed in cutaneous T-cell lymphoma, and its targeting promotes potent killing of malignant cells. Blood Adv. 2020;4(20):5203-5214. doi:10.1182/bloodadvances.2020002395
6. U.S. Department of Health and Human Services: Centers for Disease Control and Prevention. Guidelines for Safe WorkPractices in Human and Animal Medical Diagnostic Laboratories. Supplement / Vol. 61, January 6, 2012. https://www.cdc.gov/mmwr/pdf/other/su6101.pdf

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 jusque Verwendbar bis	LOT — Lot Number Code du lot Chargenbezeichnung

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