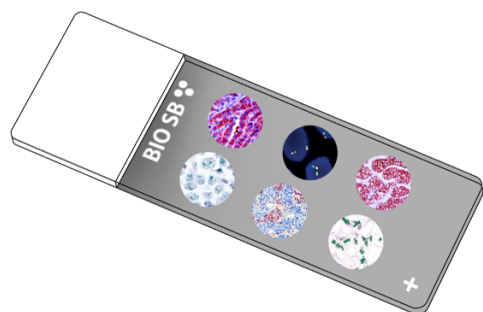


TIGIT Control Slides



Intended Use

For In Vitro Diagnostic Use.

Summary and Explanation

TIGIT (T cell immunoreceptor with Ig and ITIM domains) is an immune receptor present on some T cells and Natural Killer Cells. It is also identified as WUCAM and Vstm3. TIGIT could bind to CD155 on dendritic cells, macrophages, and other immune cells with high affinity, and also to CD112 (PVRL2) with lower affinity. Research has shown that TIGIT-Fc fusion protein could interact with PVR on dendritic cells and increase its IL-10 secretion level/decrease its IL-12 secretion level under LPS stimulation, and also inhibit T cell activation in vivo. TIGIT's inhibition of NK cytotoxicity can be blocked by antibodies against its interaction with PVR, and the activity is directed through its ITIM domain. TIGIT is expressed on regulatory T cells (Tregs) and on activated CD4+ T, CD8+ T, and NK cells. TIGIT and PD-1 has been shown to be over-expressed on tumor antigen-specific (TA-specific) CD8+ T cells and CD8+ tumor infiltrating lymphocytes (TILs) from individuals with melanoma. Blockade of TIGIT and PD-1 led to increased cell proliferation, cytokine production, and degranulation of TA-specific CD8+ T cells and TIL CD8+ T cells, therefore it can be considered an immune checkpoint. Co-blockade of TIGIT and PD-1 pathways elicits tumor rejection in preclinical murine models.

Presentation

Five slides of TIGIT positive tissues, each mounted on Hydrophilic Plus Slides, provided in a plastic mailer.

<i>Catalog No.</i>	<i>Quantity</i>
BSB-9410-CS	5 slides
BSB-3751-CS	5 slides

Storage Store at 20-25°C

Precautions

1. For professional users only. Results should be interpreted by a qualified medical professional.
2. 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. 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 package label.

IHC Protocol

1. 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).

2. 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.

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

Abbreviated IF Protocol

Step	Incubation Time
Rinse slides in IF wash buffer	5 minutes
Drain and wipe excess IF wash buffer off slide	
Conduct remaining steps in the dark	
Apply Antibody	30-60 minutes
Rinse with 3 changes of IF wash buffer	3x15 minutes each
Coverslip with IF mounting medium	

Mounting Protocols

For detailed instructions using biodegradable permanent mounting media such as XyGreen PermaMunter (BSB 0169-0174) or organic solvent based resin such as PermaMunter (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. Yu X, Harden K, Gonzalez LC, et al. The surface protein TIGIT suppresses T cell activation by promoting the generation of mature immunoregulatory dendritic cells. *Nat Immunol.* 2009;10(1):48-57. doi:10.1038/ni.16742. Levin SD, Taft DW, Brandt CS, et al. Vstm3 is a member of the CD28 family and an important modulator of T-cell function. *Eur J Immunol.* 2011;41(4):902-915. doi:10.1002/eji.2010411363. Chauvin JM, Pagliano O, Fourcade J, et al. TIGIT and PD-1 impair tumor antigen-specific CD8⁺ T cells in melanoma patients. *J Clin Invest.* 2015;125(5):2046-2058. doi:10.1172/JCI804454. Johnston RJ, Comps-Agrar L, Hackney J, et al. The immunoreceptor TIGIT regulates antitumor and antiviral CD8(+) T cell effector function. *Cancer Cell.* 2014;26(6):923-937. doi:10.1016/j.ccell.2014.10.0185. Argast G, Cancilla B, Cattaruzza F, et al. Abstract 5627: Anti-TIGIT biomarker study: Inhibition of TIGIT induces loss of Tregs from tumors and requires effector function for tumor growth inhibition. *Cancer Research.* 2018;78:5627-5627. doi:10.1158/1538-7445.AM2018-56276. 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	QAdvis EAR AB Ideon Science Park Scheelevägen 17 SE-223 70 Lund, Sweden	 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