

# B7H3/CD276

## Control Slides



### Intended Use

For In Vitro Diagnostic Use.

### Summary and Explanation

B7-H3, also known as CD276, is a human protein encoded by the CD276 gene. The protein encoded by this gene belongs to the immunoglobulin superfamily and thought to participate in the regulation of T-cell-mediated immune response. Studies show that while the transcript of this gene is ubiquitously expressed in normal tissues and solid tumors, the protein is preferentially expressed only in tumor tissues, such as melanoma, prostate cancer, and pancreatic cancer. B7-H3 mRNA is not detectable in peripheral blood mononuclear cells, although it is found in various normal tissues and in several tumor cell lines. Expression of B7-H3 protein, however, can be induced on dendritic cells (DCs) and monocytes by inflammatory cytokines. Soluble B7-H3 protein binds a putative counter-receptor on activated T cells that is distinct from CD28, cytotoxic T lymphocyte antigen 4 (CTLA-4), inducible costimulator (ICOS) and PD-1. B7-H3 costimulates proliferation of both CD4+ and CD8+ T cells, enhances the induction of cytotoxic T cells and selectively stimulates interferon gamma (IFN-gamma) production in the presence of T cell receptor signaling.

Recently, B7-H3 expression has been reported in several human cancers indicating an additional function of B7-H3 as a regulator of antitumor immunity. However, its precise physiologic role is still elusive, because both stimulatory and inhibitory capacities have been demonstrated. B7H3 has been shown in recent years to be of clinical significance in different types of cancer. In some tumor types high expression of B7-H3 has been linked to a poor prognosis, whereas in other cancers the opposite effect has been observed. Taken together, the precise role of B7-H3 in tumor immunity is unclear and further research is needed. Another aspect of B7-H3, that so far has received little interest, is its role in non-immunological systems. It has been demonstrated that knockdown of B7-H3 in melanoma and breast cancer cells results in both increased chemosensitivity and decreased metastatic potential, which has been observed in both in vitro and in vivo experiments.

### Presentation

Five slides of B7H3/CD276 positive tissues, each mounted on Hydrophilic Plus Slides, provided in a plastic mailer.

Catalog No.	Quantity
BSB-9024-CS	5 slides
BSB 2816	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

- Entrez Gene: CD276 CD276 molecule.
- Chapoval, A.I. et al. B7-H3: a costimulatory molecule for T cell activation and IFN- gamma production. Nat Immunol. 2001 Mar;2(3):269-74.
- Loos M, et al. B7-h3 and its role in antitumor immunity. Clin Dev Immunol. 2010; 2010: 683875. Published online 2010 Nov 28. doi: 10.1155/2010/683875
- Nygren MK, et al. B7-H3 and its relevance in cancer; immunological and non-immunological perspectives. Front Biosci (Elite Ed). 2011 Jun 1;3:989-93.
- 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  
<https://www.cdc.gov/mmwr/pdf/other/su6101.pdf>

### Symbol Key / Légende des symboles/Erläuterung der Symbole

<b>EC</b> <b>REP</b>	QAdvis EAR AB Ideon Science Park Scheelevägen 17 SE-223 70 Lund, Sweden	 Storage Temperature Limites de température Zulässiger Temperaturbereich	 Manufacturer Fabricant Hersteller	<b>REF</b>	Catalog Number Référence du catalogue Bestellnummer
<b>IVD</b>	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	<b>LOT</b>	Lot Number Code du lot Chargenbezeichnung