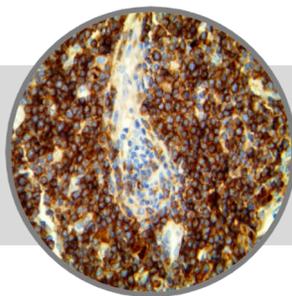


PD-L1/CD274, RMAb

Clone: 28-8

Rabbit Monoclonal

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Inset: IHC PD-L1/CD274 on a FFPE CSL/SLL Lymphoma Tissue

Intended Use

For Research Use Only.

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

Recombinant full-length protein corresponding to Human PD-L1 (extracellular).

Summary and Explanation

Programmed death-ligand 1 (PD-L1) also known as CD274 or B7 homolog 1 (B7-H1) is a protein that in humans is encoded by the *CD274* gene. Programmed death-ligand 1 (PD-L1) is a 40 kDa type 1 transmembrane protein that has been speculated to play a major role in suppressing the immune system during events such as pregnancy, tissue allografts, autoimmune disease and other diseases.

The upregulation of PD-L1 may allow cancers to evade the host immune system. An analysis of tumor specimens from patients with renal cell carcinoma found that high tumor expression of PD-L1 was associated with increased tumor aggressiveness and a 4.5-fold increased risk of death. Ovarian cancer patients with higher expression of PDL1 had a significantly poorer prognosis than those with lower expression. PD-L1 expression correlated inversely with intraepithelial CD8+ T-lymphocyte count, suggesting that PD-L1 on tumor cells may suppress antitumor CD8+ T cells. The PD1/PD-L1 interaction is implicated in autoimmunity from several lines of evidence. In humans, PD-L1 was found to have altered expression in pediatric patients with Systemic lupus erythematosus. Studying isolated PBMC from healthy children, immature myeloid dendritic cells and monocytes expressed little PD-L1 at initial isolation, but spontaneously up-regulated PD-L1 by 24 hours. In contrast, both mDC and monocytes from patients with active SLE failed to upregulate PD-L1 over a 5-day time course, expressing this protein only during disease remissions

Antibody Type	Rabbit Monoclonal	Clone	28-8
Isotype	IgG	Reactivity	Paraffin, Frozen
Localization	Membranous	Control	Tonsil, Placenta, Lymphoblastic Lymphoma
Species Reactivity		Human	

Presentation

Anti PD-L1/CD274 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.	Antibody Type	Dilution	Volume/Qty
BSB-2371-3	Tinto Predilute	Ready-to-Use	3.0 mL
BSB-2371-7	Tinto Predilute	Ready-to-Use	7.0 mL
BSB-2371-15	Tinto Predilute	Ready-to-Use	15.0 mL
BSB-2371-01	Concentrate	1:25 - 1:100	0.1 mL
BSB-2371-05	Concentrate	1:25 - 1:100	0.5 mL
BSB-2371-1	Concentrate	1:25 - 1:100	1.0 mL

Control Slides Available

Catalog No.	Quantity
BSB-2371-CS	5 slides

Precautions

1. For professional users only. Results should be interpreted by a qualified medical professional.
2. This product contains <0.1% sodium azide (NaN₃) as a preservative. Ensure proper handling procedures are used with this reagent.
3. Always wear personal protective equipment such as laboratory coat, goggles and gloves when handling reagents.
4. Dispose of unused solution with copious amount 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).

Storage Store at 2-8°C

Stability

This product is stable up to the expiration date on the product label. 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 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 for labeling acetone-fixed frozen sections and acetone-fixed cell preparations.

Staining Procedure

- Cut and mount 3-5 micron formalin-fixed paraffin-embedded tissues on positively charged slides such as Bio SB Hydrophilic Plus Slides (BSB 7028).
- Air dry for 2 hours at 58° C.
- Deparaffinize, dehydrate and rehydrate tissues.
- 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).
- Any of three heating methods may be used:
 - 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.
 - 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.
 - 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.
- After heat treatment, transfer slides in ImmunoDNA Retriever with Citrate or EDTA to room temperature and let stand for 15-20 minutes.
- For manual staining, perform antibody incubation at ambient temperature. For automated staining methods, perform antibody incubation according to instrument manufacturer's instructions.
- Wash slides with ImmunoDNA washer or DI water.
- 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

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 using positive and negative controls. Results should be interpreted by a qualified medical professional.

References

- Alsaab HO, et al. PD-1 and PD-L1 Checkpoint Signaling Inhibition for Cancer Immunotherapy: Mechanism, Combinations, and Clinical Outcome". *Frontiers in Pharmacology*. 2017; 8: 561.
- Thompson RH, et al. Costimulatory B7-H1 in renal cell carcinoma patients: Indicator of tumor aggressiveness and potential therapeutic target". *Proceedings of the National Academy of Sciences of the United States of America*. 2004; 101 (49): 17174-9.
- Mozaffarian, N, et al. Active systemic lupus erythematosus is associated with failure of antigen-presenting cells to express programmed death ligand-1. *Rheumatology*, 2008; 47 (9): 1335-1341.
- 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

	Storage Temperature Limites de température Zulässiger Temperaturbereich		Manufacturer Fabricant Hersteller	REF	Catalog Number Référence du catalogue Bestellnummer
	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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