abcam

Product datasheet

Anti-PD-L1 antibody [28-8] - BSA and Azide free ab228413

敲除验证 重组 RabMAb

1 Abreviews 17 图像

概述

产品名称	Anti-PD-L1抗体[28-8] - BSA and Azide free
描述	兔单克隆抗体[28-8] to PD-L1 - BSA and Azide free
宿主	Rabbit
经测试应用	**适用于:** WB, ICC/IF, Flow Cyt, IHC-P
种属反应性	**与反应:** Human
免疫原	Fusion protein. This information is proprietary to Abcam and/or its suppliers.
阳性对照	Tissue: Human tonsil, head and neck squamous cell carcinoma and placenta tissues; L2987 cell line. Cell Lines: Positives: B-CPAP (high), ES-2 (medium), HCC70 (low), CHO-PDL1, U-87 MG For additional information - please refer to this publication:Programmed death-ligand 1 (PD-L1) expression in various tumor types - http://www.immunotherapyofcancer.org/content/1/S1/P53 IHC-Fr: Frozen human tonsil tissue sections
常规说明	ab228413 is the carrier-free version of abab205921. **Additional information on positive controls:** Tissue: Tonsil- with hyperreactive changes Note: Tonsil Specimens- is recommended to screen several hyper-reactive tonsils to find those with highest expression of PD-L1 in crypt epithelium, macrophages homing the germinal centers and interfollicular mononuclear leukocytes. Tumor tissues- prescreened for positive tumor and inflammatory infiltrates Note: Tumor Specimens- PD-L1 expression varies by tumor type so screening is recommended to find positive and negative tumor controls. Refer to web link publication below to find some suggested tumor types. Many tumor specimens have some inflammatory macrophages and mononuclear leukocytes. Look for specimens with high numbers of these cells Cell Lines: Positives: B-CPAP- high, ES-2- medium, HCC70 - low **For IHC on FFPE tissues, antigen retrieval buffer (ab208572)** and **IHC detection kit HRP/DAB (ab209101)** is recommended. **For primary negative control, isotype control**, **RabMAb negative control antibody (ab172730)** is recommended. **For negative control sample, use cell line COLO205, see ab95363.** **For PD-L1 protein, see ab167713** **Recommended protocols:**

For IHC usage on FFPE tissues, the following antigen solution is recommended with clone 28-8 - Universal HIER antigen retrieval reagent (**ab208572**)

Western blot usage
For clone 28-8, it is recommended to use Odyssey system. This system has the advantages of a wider dynamic range and less background than chemiluminescence.

Anti-PD-L1 antibody [28-8] has been used as detector antibody in **Human PD-L1 SimpleStep ELISA® kit (ab214565)**.

Our **carrier-free** antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency.

This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our **conjugation kits** for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc.

This product is a recombinant monoclonal antibody, which offers several advantages including:
- High batch-to-batch consistency and reproducibility
- Improved sensitivity and specificity
- Long-term security of supply
- Animal-free production

For more information **see here**.

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb® patents**.

性能

形式	Liquid
存放说明	Shipped at 4°C. Store at +4°C. Do Not Freeze.
存储溶液	pH: 7.20 Constituent: 100% PBS
无载体	**是**
纯度	Protein A purified
克隆	单克隆
克隆编号	28-8
同种型	IgG

应用

The Abpromise guarantee	**Abpromise™承诺保证使用**ab228413**于以下的**经测试应用

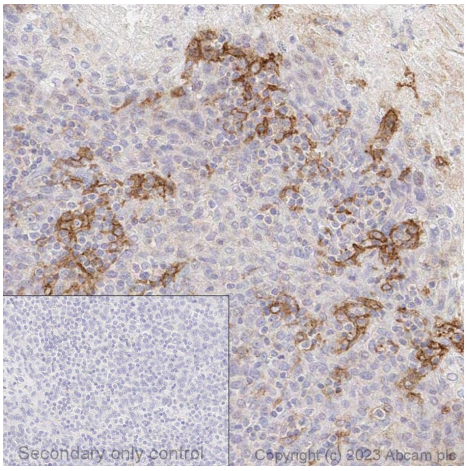
“应用说明”部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
WB		Use at an assay dependent concentration. Predicted molecular weight: 33 kDa.
ICC/IF		Use at an assay dependent concentration.
Flow Cyt		Use at an assay dependent concentration. **ab172730** - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
IHC-P	★☆☆☆☆ (1)	Use at an assay dependent concentration. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. For antigen buffer for FFPE tissue, it is recommended to use Universal HIER antigen retrieval reagent (**ab208572**).

靶标

功能	Involved in the costimulatory signal, essential for T-cell proliferation and production of IL10 and IFNG, in an IL2-dependent and a PDCD1-independent manner. Interaction with PDCD1 inhibits T-cell proliferation and cytokine production.
组织特异性	Highly expressed in the heart, skeletal muscle, placenta and lung. Weakly expressed in the thymus, spleen, kidney and liver. Expressed on activated T- and B-cells, dendritic cells, keratinocytes and monocytes.
序列相似性	Belongs to the immunoglobulin superfamily. BTN/MOG family. Contains 1 Ig-like C2-type (immunoglobulin-like) domain. Contains 1 Ig-like V-type (immunoglobulin-like) domain.
细胞定位	Cell membrane and Endomembrane system.

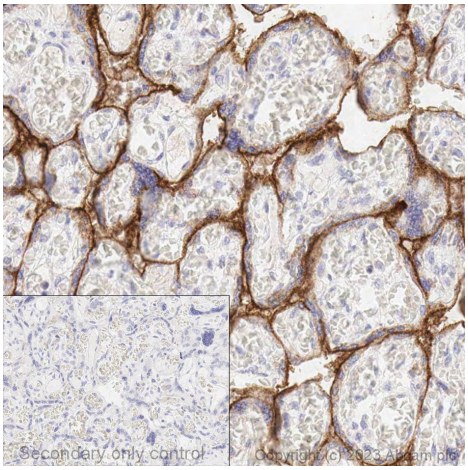
图片



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PD-L1 antibody [28-8] - BSA and Azide free (ab228413)

Immunohistochemical analysis of formalin-fixed paraffin-embedded human tonsil labelling PD-L1 with **ab205921** at a dilution of 2µg/ml. The immunostaining was performed on a Ventana DISCOVERY ULTRA (Roche Tissue Diagnostics) instrument with an OptiView DAB IHC Detection Kit. Heat mediated antigen retrieval was conducted for 32min with ULTRA cell conditioning solution (CC1 pH8.5). **ab205921** anti PD-L1 antibody was incubated at 37°C for 16min. Sections were counterstained is with Hematoxylin II. Image inset shows absence of staining in secondary antibody only control.

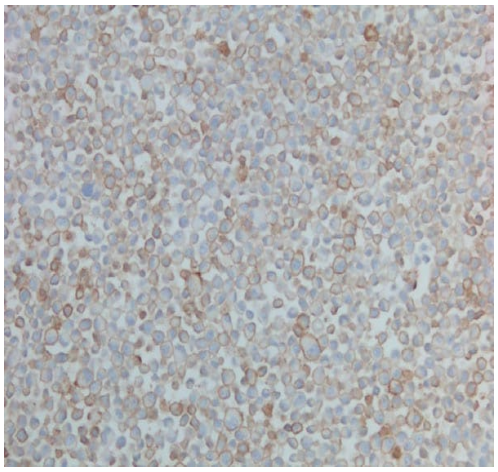
This data was developed using **ab205921**, the same antibody clone in a different buffer formulation.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PD-L1 antibody [28-8] - BSA and Azide free (ab228413)

Immunohistochemical analysis of formalin-fixed paraffin-embedded human placenta labelling PD-L1 with **ab205921** at a dilution of 2µg/ml. The immunostaining was performed on a Ventana DISCOVERY ULTRA (Roche Tissue Diagnostics) instrument with an OptiView DAB IHC Detection Kit. Heat mediated antigen retrieval was conducted for 32min with ULTRA cell conditioning solution (CC1 pH8.5). **ab205921** anti PD-L1 antibody was incubated at 37°C for 16min. Sections were counterstained is with Hematoxylin II. Image inset shows absence of staining in secondary antibody only control.

This data was developed using **ab205921**, the same antibody clone in a different buffer formulation.

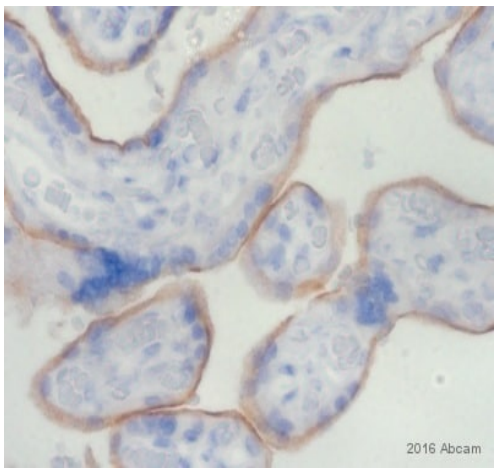


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PD-L1 antibody [28-8] - BSA and Azide free (ab228413)

Immunohistochemical analysis of formalin-fixed, paraffin-embedded L2987 (Human lung adenocarcinoma cell line with endogenous PD-L1 expression) cells labeling PD-L1 with **ab205921** at 2 µg/ml. Counterstained with Hematoxylin.

For antigen retrival buffer, Universal HIER antigen retrieval reagent (**ab208572**) was used.
For IHC detection kit, Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**) is recommended.

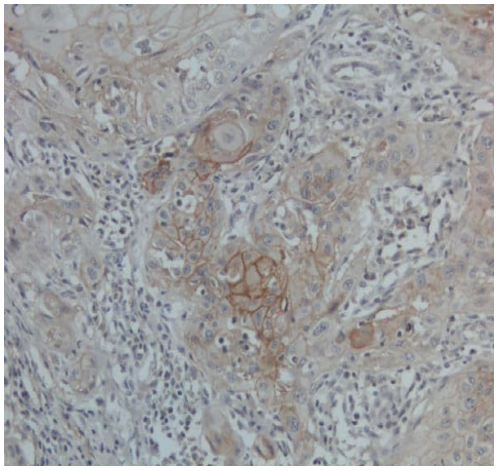
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab205921**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PD-L1 antibody [28-8] - BSA and Azide free (ab228413)

Paraformaldehyde-fixed, paraffin-embedded human placenta tissue stained for PD-L1 using **ab205921** at 1/100 dilution in immunohistochemical analysis.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab205921**).

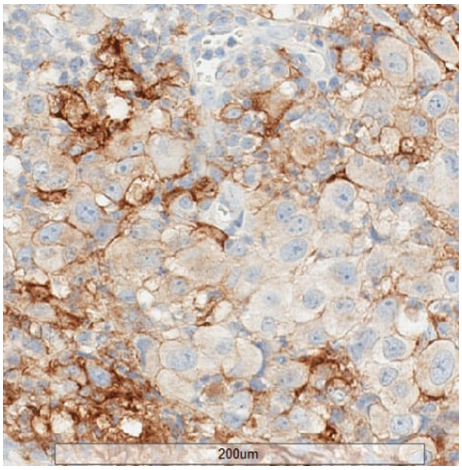


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PD-L1 antibody [28-8] - BSA and Azide free (ab228413)

Immunohistochemical analysis of formalin-fixed, paraffin-embedded Human head and neck squamous cell carcinoma tissue labeling PD-L1 with **ab205921** at 2 µg/ml. Counterstained with Hematoxylin.

For antigen retrival buffer, Universal HIER antigen retrieval reagent (**ab208572**) was used.
For IHC detection kit, Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**) is recommended.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab205921**).

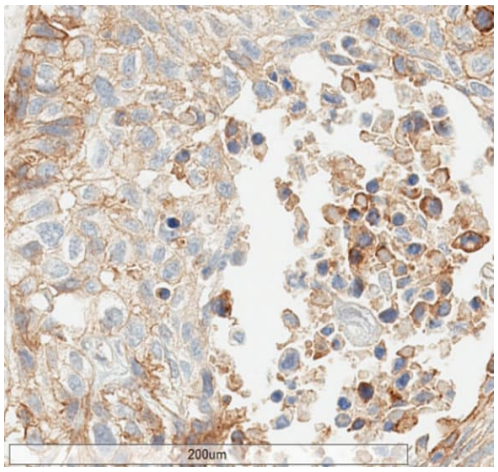


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PD-L1 antibody [28-8] - BSA and Azide free (ab228413)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human melanoma tissue labelling PD-L1 with **ab205921** on Ventana Ultra. Tumor cells and immune cells show PD-L1 positive plasma membrane staining.

For antigen retrival buffer, Universal HIER antigen retrieval reagent (**ab208572**) was used.
For IHC detection kit, Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**) is recommended.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab205921**).

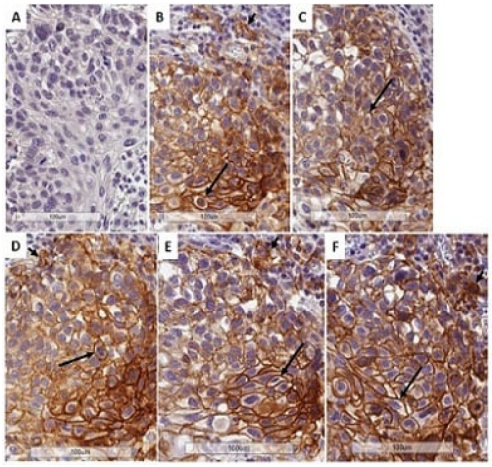


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PD-L1 antibody [28-8] - BSA and Azide free (ab228413)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human non-small cell lung cancer tissue labelling PD-L1 with **ab205921**. Staining can be seen in tumor associated macrophages and tumor cells.

For antigen retrival buffer, Universal HIER antigen retrieval reagent (**ab208572**) was used.
For IHC detection kit, Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**) is recommended.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab205921**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PD-L1 antibody [28-8] - BSA and Azide free (ab228413)

Immunohistochemical analysis of Human Lung NSCLC with **ab205921** at 2 µg/ml.
High power view
A) Rabbit IgG, 5 µg/mL. No staining
B) Anti PD-L1, 2 µg/mL (**ab205921** batches 1)
C) Anti PD-L1, 2 µg/mL (**ab205921** batches 3)
D) Anti PD-L1, 2 µg/mL (**ab205921** batches 4)
E) Anti PD-L1, 2 µg/mL (**ab205921** batches 5)
F) Anti PD-L1, 2 µg/mL (**ab205921** batches 6)

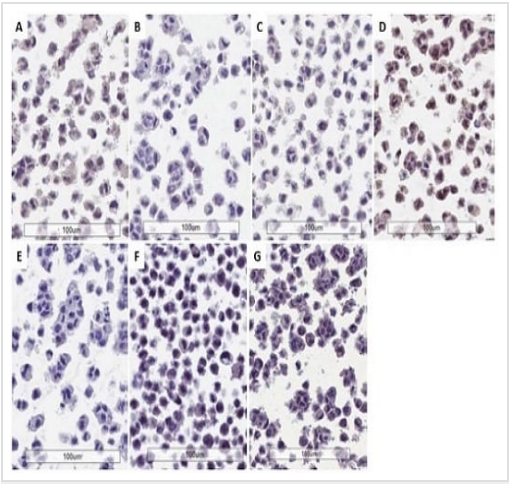
All batches/lots (1,3,4,5,6) showed consistent results.

Note linear and complete or partial (arrows) PD-L1 staining of tumor cells. Tumor associated immune cells localized over the tumor margin exhibit positive plasma membrane staining (small arrows).

For recommended Immunohistochemistry (IHC) protocol, please refer to the protocol book in the protocol section and/or **here (downloadable copy).**

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab205921**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PD-L1 antibody [28-8] - BSA and Azide free (ab228413)

Immunohistochemical analysis of CHO Parental cells with **ab205921** at 2 µg/ml.
High power view
A) Rabbit IgG, 5 µg/mL. No staining
B) Anti PD-L1, 2 µg/mL (**ab205921** batches 1)
C) Anti PD-L1, 2 µg/mL (**ab205921** batches 3)
D) Anti PD-L1, 2 µg/mL (**ab205921** batches 4)
E) Anti PD-L1, 2 µg/mL (**ab205921** batches 5)
F) Anti PD-L1, 2 µg/mL (**ab205921** batches 6)
G) Anti PD-L1, 2 µg/mL (**ab205921** batches 7)

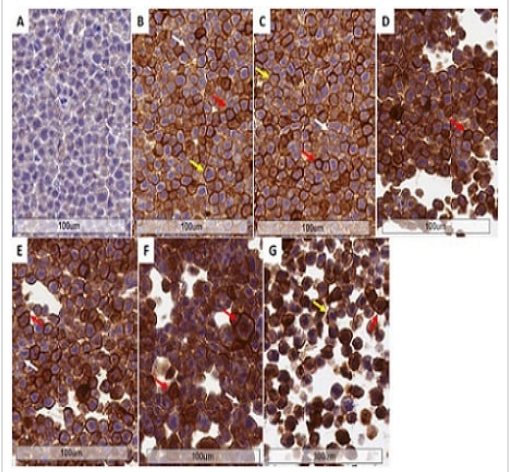
All batches/lots (1,3,4,5,6,7) showed consistent results.

Note absence of PD-L1 expression in CHO parental cells.

For recommended Immunohistochemistry (IHC) protocol, please refer to the protocol book in the protocol section and/or **here (downloadable copy).**

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab205921**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PD-L1 antibody [28-8] - BSA and Azide free (ab228413)

Immunohistochemical analysis of CHO PD-L1 cells with **ab205921** at 2 µg/ml.
High power view
A) Rabbit IgG, 5 µg/mL. No staining
B) Anti PD-L1, 2 µg/mL (**ab205921** batches 1)
C) Anti PD-L1, 2 µg/mL (**ab205921** batches 3)
D) Anti PD-L1, 2 µg/mL (**ab205921** batches 4)
E) Anti PD-L1, 2 µg/mL (**ab205921** batches 5)
F) Anti PD-L1, 2 µg/mL (**ab205921** batches 6)
G) Anti PD-L1, 2 µg/mL (**ab205921** batches 7)

All batches/lots (1,3,4,5,6,7) showed consistent results.

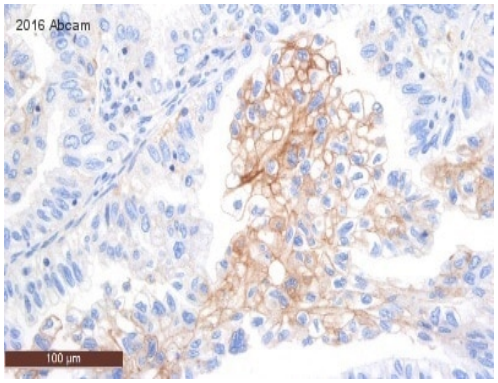
Note strong, moderate, and weak (red, yellow, and white arrows respectively) plasma membrane staining of CHO PD-L1 transfected

cells

For recommended Immunohistochemistry (IHC) protocol, please refer to the protocol book in the protocol section and/or **here (downloadable copy).**

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab205921**).

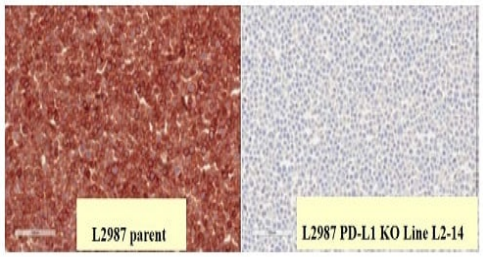


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PD-L1 antibody [28-8] - BSA and Azide free (ab228413)

Immunohistochemical staining of PD-L1 in formalin fixed, paraffin embedded human non-squamous non-small cell lung cancer (NSQ-NSCLC) using **ab205921** at a dilution of 1/400, incubated for an hour at room temperature. Heat mediated antigen retrieval was carried out in low pH buffer and the sample was blocked with peroxidase blocking buffer for 3 minutes.

This image was courteously provided by Dr. Kai Schmitt from the Institute of Pathology, Saarbrücken-Rastpfuhl.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab205921**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PD-L1 antibody [28-8] - BSA and Azide free (ab228413)

Ab205921 specificity testing by Immunohistochemistry (KO testing): Loss of detection on KO Cells

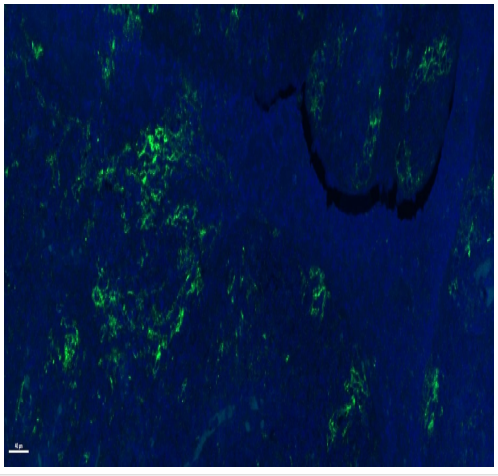
Strong IHC detection with anti-PD-L1 (**ab205921**, clone 28-8) is seen in human lung adenocarcinoma tumor cell line L2987. PDL1 gene was edited in L2987 cells using TALEN constructs targeting exon4 of human PD-L1, transcript variant 1 (NM_014143.3) and complete knock out (K.O) confirmed by deep sequencing in clone L2-14. IHC detection is completely eliminated in the L2987 L2-14 K.O. cell line.

For recommended Immunohistochemistry (IHC) protocol, please refer to the protocol book in the protocol section and/or **here (downloadable copy).**

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

This data was developed using the same antibody clone in a

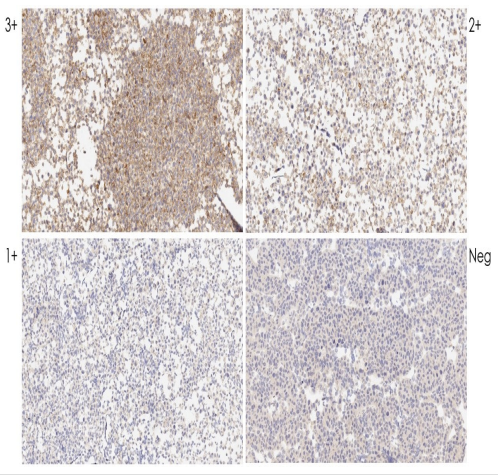
different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab205921**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PD-L1 antibody [28-8] - BSA and Azide free (ab228413)

Anti-PD-L1 antibody [28-8] (**ab205921**) Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human tonsil tissue labelling PD-L1 with **ab205921** at a dilution of 1:400. Heat mediated antigen retrieval was performed using AR9 antigen retrieval solution, and microwave treatment for 15 min at 20% power. Anti-Rabbit/Mouse HRP polymer (PerkinElmer Opal Polymer HRP Ms Plus Rb) was used as secondary antibody. Opal tyramide amplification was performed using Opal 520 fluorophore. Counterstained with DAPI stain. Image scanned with Vectra 3.0 and analyzed via Phenochart software. This image was courteously provided by Dr. Houssein Abdul Sater, Georgia Cancer Center.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab205921**).

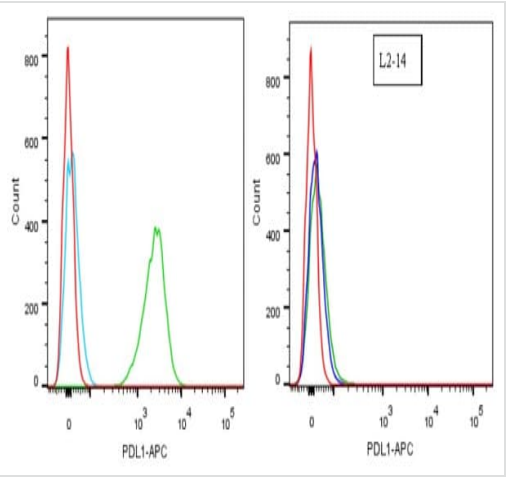


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PD-L1 antibody [28-8] - BSA and Azide free (ab228413)

IHC image of **ab205921** staining PD-L1 in PD-L1 Dynamic Range Analyte Control formalin fixed paraffin embedded cell lines (**HistoCyte Laboratories**), performed on a Leica BOND RX (Polymer Refine kit). The section was pre-treated using heat mediated antigen retrieval with EDTA buffer (pH9, epitope retrieval solution 2) for 30 mins at 98°C. The section was then incubated with **ab205921**, 5µg/ml working concentration, for 60 mins at room temperature and detected using an HRP conjugated compact polymer system for 8 minutes at room temperature. DAB was used as the chromogen for 10 minutes at room temperature. The section was then counterstained with hematoxylin, blued, dehydrated, cleared and mounted with DPX.

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab205921**).



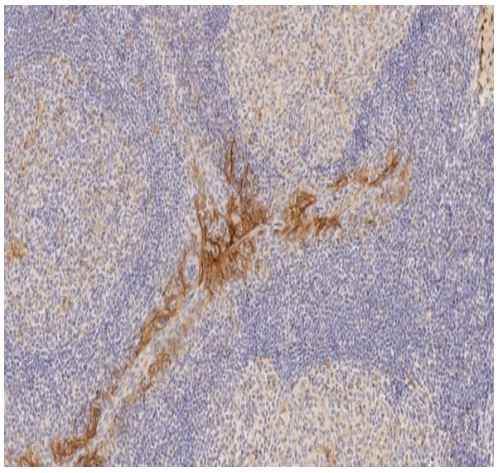
Flow Cytometry - Anti-PD-L1 antibody [28-8] - BSA and Azide free (ab228413)

ab205921 specificity testing by Flow Cytometry (KO testing): Loss of detection on KO cells.
Strong detection with anti-PD-L1 (**ab205921**, clone 28-8) TALEN constructs targeting exon4 of human PD-L1, transcript variant 1 (NM_014143.3) and complete knock out (K.O) confirmed by deep sequencing in clone L2-14. Cell surface staining is almost completely eliminated in the L2987 L2-14 KO cell line.

For recommended Flow Cytometry (Flow Cyt) protocol, please refer to the protocol book in the protocol section and/or **here (downloadable copy).**

Alexa Fluor® 488 (**ab209959**) and Alexa Fluor® 647 (**ab209960**) conjugated versions are available for this clone.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab205921**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PD-L1 antibody [28-8] - BSA and Azide free (ab228413)

IHC image of **ab205921** staining PD-L1 in human tonsil formalin fixed paraffin embedded tissue sections*, performed on a Leica BOND RX (Polymer Refine kit). The section was pre-treated using heat mediated antigen retrieval with EDTA buffer (pH9, epitope retrieval solution 2) for 30 mins at 98°C. The section was then incubated with **ab205921**, 5µg/ml working concentration, for 60 mins at room temperature and detected using an HRP conjugated compact polymer system for 8 minutes at room temperature. DAB was used as the chromogen for 10 minutes at room temperature. The section was then counterstained with hematoxylin, blued, dehydrated, cleared and mounted with DPX.

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.

*Tissue obtained from the Human Research Tissue Bank, supported by the NIHR Cambridge Biomedical Research Centre

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab205921**).

Anti-PD-L1 antibody [28-8] - BSA and Azide free

(ab228413)

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