abcam

Product datasheet

Anti-PD-L1 antibody [73-10] - Low endotoxin, Azide free ab246698

יובעדער RabMAb

画像数 20

製品の概要

製品名 Anti-PD-L1 antibody [73-10] - Low endotoxin, Azide free

製品の詳細 Rabbit monoclonal [73-10] to PD-L1 - Low endotoxin, Azide free

由来種 Rabbit

アプリケーション 適用あり: WB, ICC/IF, IHC-P, IP, Flow Cyt (Intra)

種交差性 交差種: Human, Recombinant fragment

免疫原 Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

ポジティブ・コントロール IHC-P: Human placenta, lung carcinoma and tonsil tissue. ICC/IF: CHO-PD-L1 Cells IP: NCI-

H1975 cells Flow Cyt (intra): CHO-PD-L1

特記事項 ab246698 is the carrier-free version of ab226766.

Clone 73-10 is also known as clone MKP1A07310.

Clone 73-10 has been tested within Blueprint Phase 2 project.

See here for more details.

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 cellbased 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 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.

Our <u>Low endotoxin, azide-free formats</u> have low endotoxin level (≤ 1 EU/ml, determined by the LAL assay) and are free from azide, to achieve consistent experimental results in functional assays.

製品の特性

製品の状態 Liquid

保存方法 Shipped at 4°C. Store at +4°C. Do Not Freeze.

バッファー pH: 7.2

Constituent: PBS

キャリア・フリー はい

精製度 Protein A purified

特記事項(精製) Endotoxin level is less than 1 EU/ml as determined by the TAL test.

ポリ/モノ モノクローナル

クローン名 73-10 **Pイソタイプ I**gG

アプリケーション

The Abpromise guarantee <u>Abpromise保証は、</u>次のテスト済みアプリケーションにおけるab246698の使用に適用されます アプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
WB		Use at an assay dependent concentration. Predicted molecular weight: 33 kDa.
ICC/IF		Use at an assay dependent concentration.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.
IP		Use at an assay dependent concentration.
Flow Cyt (Intra)		Use at an assay dependent concentration.

ターゲット情報

機能 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.

組織特異性 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.

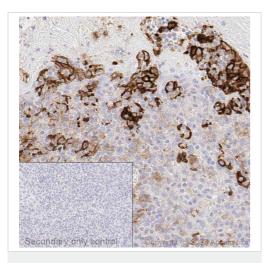
配列類似性

Belongs to the immunoglobulin superfamily. BTN/MOG family. Contains 1 lg-like C2-type (immunoglobulin-like) domain. Contains 1 lg-like V-type (immunoglobulin-like) domain.

細胞内局在

Cell membrane and Endomembrane system.

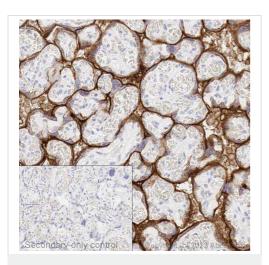
画像



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PD-L1 antibody [73-10] - Low endotoxin, Azide free (ab246698)

Immunohistochemical analysis of formalin-fixed paraffin-embedded human tonsil labelling PD-L1 with <u>ab228415</u> at a concentration of 0.1µg/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). <u>ab228415</u> anti PD-L1 antibody was incubated at 37°C for 16min. Sections were counterstained with Hematoxylin II. Image inset shows absence of staining in secondary antibody only control.

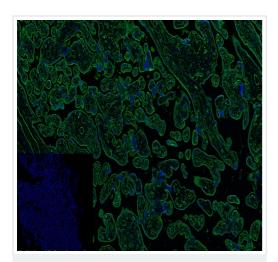
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab228415).



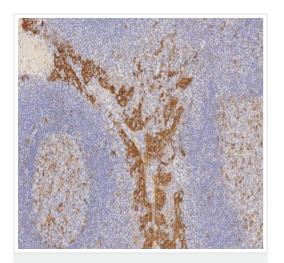
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PD-L1 antibody [73-10] - Low endotoxin, Azide free (ab246698)

Immunohistochemical analysis of formalin-fixed paraffin-embedded human placenta labelling PD-L1 with <u>ab228415</u> at a concentration of 0.1µg/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). <u>ab228415</u> anti PD-L1 antibody was incubated at 37°C for 16min. Sections were counterstained with Hematoxylin II. Image inset shows absence of staining in secondary antibody only control.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab228415).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PD-L1 antibody [73-10] - Low endotoxin, Azide free (ab246698)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PD-L1 antibody [73-10] - Low endotoxin, Azide free (ab246698)

Anti-PD-L1 antibody [73-10] (ab228415)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human tonsil tissue labelling PD-L1 with **ab228415** at a dilution of 1:2500. 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 (Vector Labs) 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 software.

This image was courteously provided by Dr. Houssein Abdul Sater, Georgia Cancer Center.

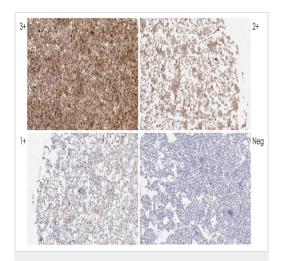
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol and sodium azide (ab228415).

IHC image of <u>ab228415</u> staining PD-L1 in human tonsil formalin fixed paraffin embedded tissue sections*, performed on a Leica BOND RX (standard Protocol F, 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 <u>ab228415</u>, 0.06µg/ml working concentration, for 15 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 image was generated using <u>ab228415</u>, the same antibody but with BSA and Azide



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PD-L1 antibody [73-10] - Low endotoxin, Azide free (ab246698)

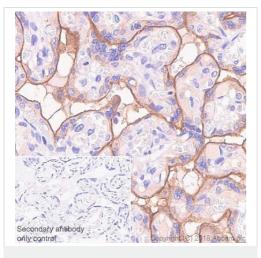
IHC image of <u>ab228415</u> staining PD-L1 in PD-L1 Dynamic Range Analyte Control formalin fixed paraffin embedded cell lines (<u>HistoCyte Laboratories</u>), performed on a Leica BOND RX (standard Protocol F, Polymer Refine kit). The section was pretreated using heat mediated antigen retrieval with EDTA buffer (pH9, epitope retrieval solution 2) for 30 mins at 98°C. The section was then incubated with <u>ab228415</u>, 0.06µg/ml working concentration, for 15 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.

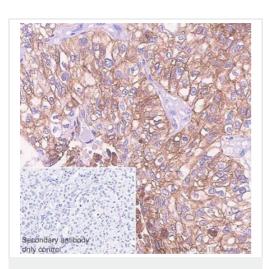
This image was generated using <u>ab228415</u>, the same antibody but with BSA and Azide

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) of Human placenta stainging PD-L1 with <u>ab228415</u> at 1/500 dilution. Heat mediated antigen retrieval was performed using Bond™ Epitope Retrieval Solution 2 (pH 9.0) for 10 mins. The section was incubated with <u>ab228415</u> for 10 mins at room temperature. The secondary antibody used was ready to use Rabbit specific IHC polymer detection kit HRP/DAB (<u>ab209101</u>). Counter stained Hematoxylin. Performed on a Leica Biosystems BOND® RX instrument.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol and sodium azide (<u>ab228415</u>).



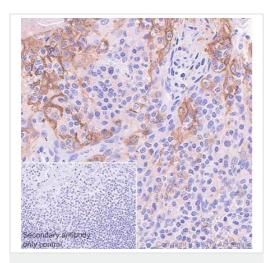
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PD-L1 antibody [73-10] - Low endotoxin, Azide free (ab246698)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PD-L1 antibody [73-10] - Low endotoxin, Azide free (ab246698)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) of Human lung carcinoma stainging PD-L1 with <u>ab228415</u> at 1/500 dilution. Heat mediated antigen retrieval was performed using Bond™ Epitope Retrieval Solution 2 (pH 9.0) for 10 mins. The section was incubated with <u>ab228415</u> for 10 mins at room temperature. The secondary antibody used was ready to use Rabbit specific IHC polymer detection kit HRP/DAB (<u>ab209101</u>). Counter stained Hematoxylin. Performed on a Leica Biosystems BOND® RX instrument.

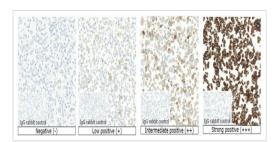
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol and sodium azide (<u>ab228415</u>).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PD-L1 antibody [73-10] - Low endotoxin, Azide free (ab246698)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) of Human tonsil stainging PD-L1 with <u>ab228415</u> at 1/500 dilution. Heat mediated antigen retrieval was performed using Bond™ Epitope Retrieval Solution 2 (pH 9.0) for 10 mins. The section was incubated with <u>ab228415</u> for 10 mins at room temperature. The secondary antibody used was ready to use Rabbit specific IHC polymer detection kit HRP/DAB (<u>ab209101</u>). Counter stained Hematoxylin. Performed on a Leica Biosystems BOND® RX instrument.

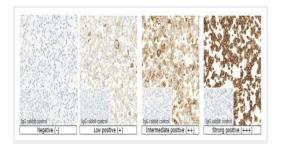
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol and sodium azide (ab228415).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PD-L1 antibody [73-10] - Low endotoxin, Azide free (ab246698)

Immunohistochemical staining of PD-L1 in formalin-fixed, paraffin embedded Formalin-fixed, paraffin-embedded reference standard with negative (-), low positive (+), intermediate positive (++) and strong positive (+++) controlled protein expressing cell lines (,CD274 (PD-L1) Expression IHC Reference Standard', catalog ID HD787, horizon) using clone 73-10 [ab228415] at a dilution of 10µg/ml. Incubate for 30 minutes at 37°C. Heat mediated antigen retrieval in sCC1 (Tris/EDTA buffer, pH 8). Signal detection with BenchMark XT from Roche/Ventana and ultraView Universal DAB Detection Kit (Code 760-500).

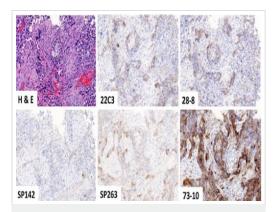
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol and sodium azide (ab228415).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PD-L1 antibody [73-10] - Low endotoxin, Azide free (ab246698)

Immunohistochemical staining of PD-L1 in formalin-fixed, paraffin embedded reference standard with negative (-), low positive (+), intermediate positive (++) and strong positive (+++) controlled protein expressing cell lines (,CD274 (PD-L1) Expression IHC Reference Standard, catalog ID HD787, horizon) using clone 73-10 [ab228415] at a dilution of 2µg/ml. Incubate for 30 minutes at room temperature. Heat mediated antigen retrieval in high pH buffer (Tris/EDTA buffer, pH 9, during 20 min at 95°C). Block sample with peroxidase blocking buffer (EnVision Flex Peroxidase-Blocking Reagent) for 5 minutes. Signal detection with Autostainer Link from Dako and EnVision Flex Kit, High pH (Code K8000).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol and sodium azide (ab228415).

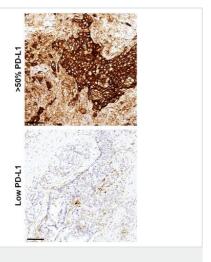


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PD-L1 antibody [73-10] - Low endotoxin, Azide free (ab246698)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) of lung cancer tissue samples. Comparing the staining PD-L1 with different monoclonal antibodies. 73-10 showed higher sensitivity to PD-L1 compared to the other clones. For further details on this image please see PubMed ID: 29800747.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol and sodium azide (ab228415).

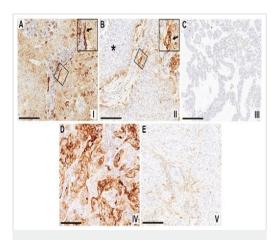
Heat mediated antigen retrieval was performed before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PD-L1 antibody [73-10] - Low endotoxin, Azide free (ab246698)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) Staining PD-L1 in human non-small cell lung cancer tissue with >50% PD-L1-positive tumor cells were compared with tissue with lower PD-L1 expression using 73-10 at 0.25µg/ml incubated for 30 minutes at room temperature. Antigen Retrieval was done with Target Retrieval Solution, high pH. Detection was done with EnVision FLEX/HRP. Hematoxylin EnVision FLEX was used as a counter stain.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol and sodium azide (ab228415).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PD-L1 antibody [73-10] - Low endotoxin, Azide free (ab246698)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) Staining PD-L1 in human non-small cell lung cancer tissue using 73-10 at 0.25µg/ml incubated for 30 minutes at room temperature. Antigen Retrieval was done with Target Retrieval Solution, high pH. Detection was done with EnVision FLEX/HRP. Hematoxylin EnVision FLEX was used as a counter stain.

A: Diffuse expression of PD-L1 (IHC) on tumor cell membranes of a squamous cell carcinoma, including central regions of trabeculae. Prominent labeling of cells in the TME compartment at the tumornest-TME interface suggesting presence of an immunological synapse (inset arrow).

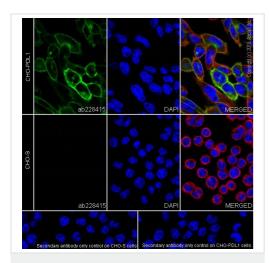
B: Patchy expression of PD-L1 in a squamous cell carcinoma at the tumor-nest-TME interface (inset arrow). Minimal to no PD-L1 expression in the trabeculae (asterisk) if compared with (**A**)

C: No to minimal PD-L1 expression in both tumor and TME compartments in an adenocarcinoma.

D: Diffuse expression of PD-L1 by tumor-nests in an adenocarcinoma with minimal TME staining.

F: TME expression only. No to minimal PD-L1 expression in tumor cells of a squamous cell carcinoma, with widespread staining in the TME compartment.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol and sodium azide (ab228415).



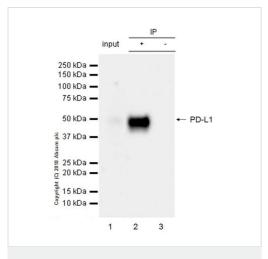
Immunocytochemistry/ Immunofluorescence - Anti-PD-L1 antibody [73-10] - Low endotoxin, Azide free (ab246698)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized CHO-PD-L1 (PD-L1 stably expressed Chinese hamster ovary epithelial cell) cells labeling PD-L1 with ab228415 at 1/200 dilution followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor[®] 488) (ab150077) secondary antibody at 1/1,000 dilution (green). Confocal image showing membranous staining on CHO-PD-L1 cells.

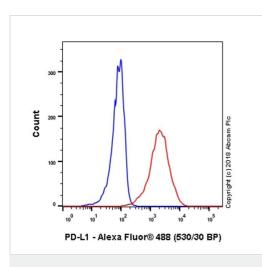
The nuclear counter stain is DAPI (blue). Tubulin is detected with anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor[®] 594) (ab195889) (red) at 1/200 dilution.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (Alexa Fluor[®] 488) (ab150077) secondary antibody at 1/1,000 dilution.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol and sodium azide (ab228415).



Immunoprecipitation - Anti-PD-L1 antibody [73-10] - Low endotoxin, Azide free (ab246698)



Flow Cytometry (Intracellular) - Anti-PD-L1 antibody [73-10] - Low endotoxin, Azide free (ab246698)

PD-L1 was immunoprecipitated from 0.35 mg of NCI-H1975 (human non-small cell lung cancer cell line) whole cell lysate with ab228415 at 1/30 dilution. Western blot was performed from the immunoprecipitate using ab228415 at 1/1,000 dilution. VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/5,000 dilution.

Lane 1: NCI-H1975 whole cell lysate 10 µg (Input).

Lane 2: <u>ab228415</u> IP in NCI-H1975 whole cell lysate (+).

Lane 3: Rabbit monoclonal IgG (<u>ab172730</u>) instead of <u>ab228415</u> in NCI-H1975 whole cell lysate (-).

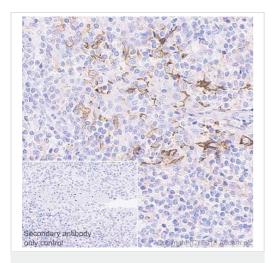
Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure time: 30 seconds.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol and sodium azide (<u>ab228415</u>).

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed, 90% methanol-permeabilized CHO-PD-L1 (PD-L1 stably expressed Chinese hamster ovary epithelial cell, Red) / CHO-S (Chinese hamster ovary epithelial cell, Blue) cell lines labeling PD-L1 with ab228415 at 1/100 dilution (red). Goat Anti-Rabbit lgG H&L (Alexa Fluor[®] 488) (ab150077) at 1/2000 dilution was used as the secondary antibody.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol and sodium azide (<u>ab228415</u>).

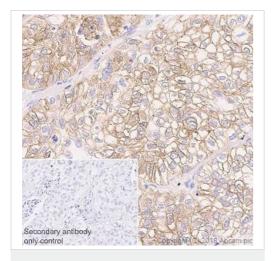


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PD-L1 antibody [73-10] - Low endotoxin, Azide free (ab246698)

Immunohistochemical analysis of paraffin-embedded human tonsil tissue labeling PD-L1 with ab228415 at 1/5000 dilution. The tissue was incubated with ab228415 at 4°C overnight, followed by Rabbit specific IHC polymer detection kit HRP/DAB (ab209101) ready to use. Cytoplasmic and membranous staining in human tonsil is observed. Counter stained with hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).

Antigen retrieval: Universal HIER antigen retrieval reagent (10X) (ab208572).

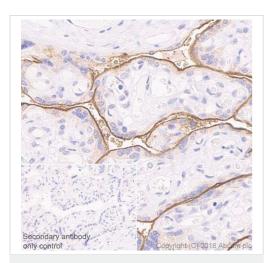


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PD-L1 antibody [73-10] - Low endotoxin, Azide free (ab246698)

Immunohistochemical analysis of paraffin-embedded human lung carcinoma tissue labeling PD-L1 with <u>ab228415</u> at 1/5000 dilution. The tissue was incubated with <u>ab228415</u> at 4°C overnight, followed by Rabbit specific IHC polymer detection kit HRP/DAB (<u>ab209101</u>) ready to use. Membranous and weakly cytoplasmic staining in human lung carcinoma (PMID: 23460533) is observed. Counter stained with hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).

Antigen retrieval: Universal HIER antigen retrieval reagent (10X) (ab208572).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PD-L1 antibody [73-10] - Low endotoxin, Azide free (ab246698)

Immunohistochemical analysis of paraffin-embedded human placenta tissue labeling PD-L1 with <u>ab228415</u> at 1/5000 dilution. The tissue was incubated with <u>ab228415</u> at 4°C overnight. followed by Rabbit specific IHC polymer detection kit HRP/DAB (<u>ab209101</u>) ready to use. Membranous and cytoplasmic staining in human placenta (PMID: 12538684) is observed. Counter stained with hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).

Antigen retrieval: Universal HIER antigen retrieval reagent (10X) (ab208572).



free (ab246698)

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