abcam

Product datasheet

Anti-LAG-3 antibody [EPR20261] - Low endotoxin, Azide free ab282638

יילעבער RabMAb

画像数8

製品の概要

製品名 Anti-LAG-3 antibody [EPR20261] - Low endotoxin, Azide free

製品の詳細 Rabbit monoclonal [EPR20261] to LAG-3 - Low endotoxin, Azide free

由来種 Rabbit

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

種交差性 交差種: Human

免疫原 Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.

ポジティブ・コントロール WB: HDLM-2 cells and Human LAG-3 Fc chimera recombinant protein (aa23-450). IHC-P:

> Human tonsil and Hodgkin's lymphoma tissues. ICC/IF: HEK-293T cells transfected with a GFPtagged LAG3 expression construct. Flow Cyt: HEK-293T transfected with a GFP-tagged human LAG-3 construct. IP: HEK-293T transfected with a GFP-tagged human LAG-3 construct whole cell

lvsate.

特記事項 ab282638 is a low endotoxin version of ab227579.

> Our Low endotoxin, azide-free formats 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.

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**.

製品の特件

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: 7.20

Constituent: 100% PBS

はい

キャリア・フリー

精製度 Protein A purified

ポリ/モノ モノクローナル

クローン名 EPR20261

アイソタイプ lgG

アプリケーション

The Abpromise guarantee Abpromise保証は、次のテスト済みアプリケーションにおけるab282638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. Detects a band of approximately 90 kDa (predicted molecular weight: 57 kDa).
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. Different mRNA expression levels of LAG3 in brain have been reported in the literature (PMID: 1692078; PMID: 12825348). In IHC, under our experimental conditions, this antibody showed no positive staining on human cerebral cortex.
Flow Cyt		Use at an assay dependent concentration.
IP		Use at an assay dependent concentration.
ICC/IF		Use at an assay dependent concentration.

ターゲット情報

機能 Involved in lymphocyte activation. Binds to HLA class-II antigens.

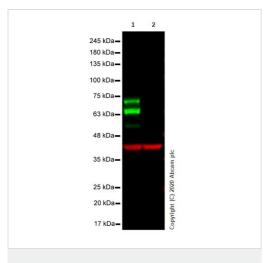
組織特異性 On cell surface of activated NK and T-lymphocytes.

配列類似性 Contains 3 lg-like C2-type (immunoglobulin-like) domains.

Contains 1 lg-like V-type (immunoglobulin-like) domain.

細胞内局在 Membrane.

画像



Western blot - Anti-LAG-3 antibody [EPR20261] - Low endotoxin, Azide free (ab282638)

All lanes : Anti-LAG-3 antibody [EPR20261] (<u>ab209236</u>) at 1/500 dilution

Lane 1 : HDLM-2 (Human Hodgkin lymphoma) whole cell lysate
Lane 2 : Jurkat (Human T cell leukemia T lymphocyte) whole cell
lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (<u>ab216773</u>) at 1/10000 dilution

Predicted band size: 57 kDa

This IHC data was generated using the same anti-LAG-3 antibody clone, EPR20261, in a different buffer formulation (ab209236).

Primary loading control and concentration: Anti-GAPDH antibody [6C5] - Loading Control (ab8245) at 1/20000 dilution

Secondary loading control and concentration: Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (ab216776) at 1/10000 dilution

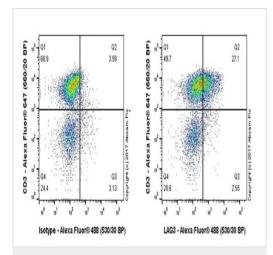
Lanes 1-2: Merged signal (red and green). Green – <u>ab209236</u> observed at 54-70 kDa. Red - loading control <u>ab8245</u> observed at 36 kDa.

<u>ab209236</u> and Anti-GAPDH antibody [6C5] - Loading Control (<u>ab8245</u>) were incubated overnight at 4°C at 1 in 500 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye® 800RCW) preadsorbed (<u>ab216773</u>) and Goat anti-Mouse lgG H&L (IRDye® 680RD) preadsorbed (<u>ab216776</u>) secondary antibodies at 1 in 10000 dilution for 1 hour at room temperature before imaging.

The expression profile observed in Jurkat is consistent with the literature (PMID: 25108024).

Negative control: Jurkat (PMID: 25108024)

Observed MW: 54-70 kDa

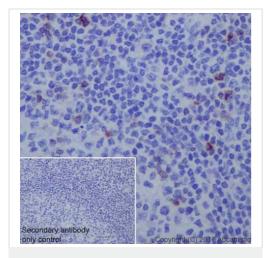


Flow Cytometry - Anti-LAG-3 antibody [EPR20261] - Low endotoxin, Azide free (ab282638)

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab209236).

Flow cytometric analysis of Human peripheral blood mononuclear cells treated with 1 μ g/mL PHA for 3 days cells with <u>ab209236</u> at 1/50 dilution (right) compared with a rabbit monoclonal lgG isotype control (<u>ab172730</u>; left). <u>ab150077</u> at 1/2000 dilution was used as the secondary antibody.

Only the CD3+ population are also positive for LAG-3. Gated on total viable cells.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-LAG-3 antibody
[EPR20261] - Low endotoxin, Azide free (ab282638)

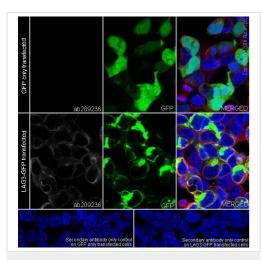
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab209236).

Immunohistochemical analysis of paraffin-embedded human tonsil tissue labeling LAG-3 with <u>ab209236</u> at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Sporadic cytoplasmic staining on immunocytes of human tonsil [PMID: 11527700; PMID: 16757686].

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) Ready to use.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunocytochemistry/ Immunofluorescence - Anti-LAG-3 antibody [EPR20261] - Low endotoxin, Azide free (ab282638)

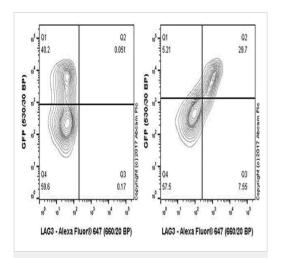
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab209236).

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HEK-293T (Human epithelial cell line from embryonic kidney) cells transfected with GFP-tagged LAG3 expression construct or GFP only, labeling LAG-3 with **ab209236** at 1/100 dilution, followed by Goat anti-rabbit lgG (Alexa Fluor[®] 647) (**ab150079**) secondary antibody at 1/1000 dilution (green).

Confocal image showing positive staining on HEK-293T cells transfected with a GFP-tagged LAG-3 expression construct.

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

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat anti-rabbit lgG (Alexa Fluor[®] 647) (ab150079) at 1/1000 dilution.

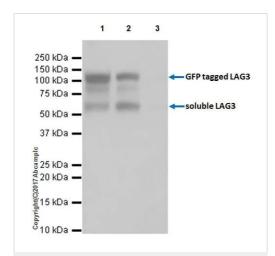


Flow Cytometry - Anti-LAG-3 antibody [EPR20261] - Low endotoxin, Azide free (ab282638)

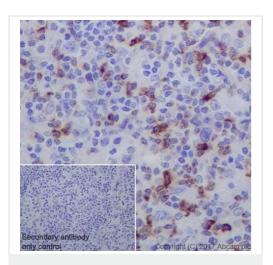
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab209236).

Flow cytometric analysis of HEK-293T (Human epithelial cell line from embryonic kidney) cells transfected with a GFP-tagged human LAG3 construct labeling LAG-3 with <u>ab209236</u> at 1/500 dilution (right) compared with a rabbit monoclonal IgG isotype control (<u>ab172730</u>; left). Goat anti rabbit IgG (Alexa Fluor[®] 647) <u>ab150079</u> at 1/2000 dilution was used as the secondary antibody.

Note: Fresh cells without fixation and permeabilization were used to perform FC testing. Only GFP positive population results in LAG3 positive staining (Q2, right panel).



Western blot - Anti-LAG-3 antibody [EPR20261] - Low endotoxin, Azide free (ab282638)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-LAG-3 antibody
[EPR20261] - Low endotoxin, Azide free (ab282638)

This IHC data was generated using the same anti-LAG-3 antibody clone, EPR20261, in a different buffer formulation (ab209236).

Immunohistochemical analysis of paraffin-embedded human tonsil Hodgkin's lymphoma labeling LAG-3 with <u>ab209236</u> at 1/1000 dilution, followed by Goat Anti-Rabbit lgG H&L (HRP) Ready to use. Cytoplasmic staining on immunocytes of the human Hodgkin's lymphoma [PMID: 11527700; PMID: 16757686].

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) Ready to use.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

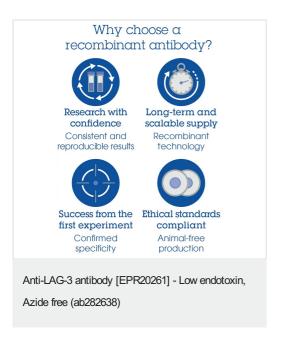
This IHC data was generated using the same anti-LAG-3 antibody clone, EPR20261, in a different buffer formulation (ab209236).

Immunohistochemical analysis of paraffin-embedded human tonsil Hodgkin's lymphoma labeling LAG-3 with <u>ab209236</u> at 1/1000 dilution, followed by Goat Anti-Rabbit lgG H&L (HRP) Ready to use. Cytoplasmic staining on immunocytes of the human Hodgkin's lymphoma [PMID: 11527700; PMID: 16757686].

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Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) Ready to use.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



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