abcam

Product datasheet

Anti-CD80 antibody [EPR1157(2)] - Low endotoxin, Azide free ab224797



リコンピナント

RabMAb

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製品の概要

特記事項

製品名 Anti-CD80 antibody [EPR1157(2)] - Low endotoxin, Azide free

製品の詳細 Rabbit monoclonal [EPR1157(2)] to CD80 - Low endotoxin, Azide free

由来種 Rabbit

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

適用なし: ICC/IF

種交差性 交差種: Human

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 WB: Raji and Ramos cell lysates. IHC-P: Human tonsil tissue. Flow Cyt (intra): Raji cells.

ab224797 is the carrier-free version of ab134120.

Our <u>carrier-free</u> 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 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.

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Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with these species. Please contact us for more information.

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

ポリモノクローナル **ウローン名** EPR1157(2)

アイソタイプ lgG

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ab224797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
Flow Cyt (Intra)		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Detects a band of approximately 60 kDa (predicted molecular weight: 33 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.

追加情報 Is unsuitable for ICC/IF.

ターゲット情報

機能 Involved in the costimulatory signal essential for T-lymphocyte activation. T-cell proliferation and

cytokine production is induced by the binding of CD28 or CTLA-4 to this receptor.

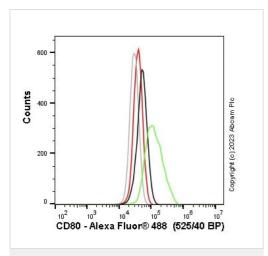
組織特異性 Expressed on activated B-cells, macrophages and dendritic cells.

配列類似性 Contains 1 lg-like C2-type (immunoglobulin-like) domain.

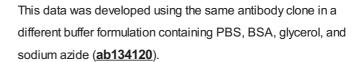
Contains 1 lg-like V-type (immunoglobulin-like) domain.

細胞内局在 Membrane.

画像



Flow Cytometry (Intracellular) - Anti-CD80 antibody [EPR1157(2)] - Low endotoxin, Azide free (ab224797)



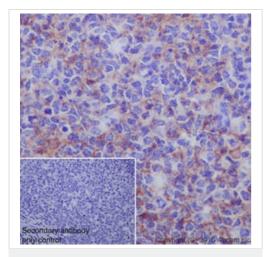
Flow cytometry overlay histogram showing wild-type Raji (green line) and CD80 knockout Raji stained with <u>ab134120</u> (red line). The cells were fixed with 4% formaldehyde (10 min) and then permeabilised with 0.1% PBS-Triton X-100 for 15 min. The cells were then incubated in 1x PBS containing 10 μ g/ml human lgG and 10% normal goat serum to block FC receptors and non-specific protein-protein interaction followed by the antibody (<u>ab134120</u>) (1x 10⁶ in 100 μ l at 0.2 μ g/ml (1/10150)) for 30min at 22°C.

The secondary antibody Goat Anti-Rabbit lgG H&L (Alexa Fluor® 488) preadsorbed was incubated at 1/4000 for 30min at 22°C

Isotype control antibody Recombinant Rabbit IgG, monoclonal [EPR25A] - Isotype Control was used at the same concentration and conditions as the primary antibody (wild-type Raji - black line, CD80 knockout Raji - grey line). Unlabelled sample was also used as a control (this line is not shown for the purpose of simplicity).

Acquisition of >5000 events were collected using a 50 mW Blue laser (488nm) and 525/40 bandpass filter.

This antibody gave a positive signal in Raji Fixed with 80% methanol (5 min) / permeabilised with 0.1% PBS-Triton X-100 for 15 min under the same conditions.

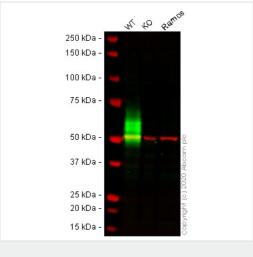


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD80 antibody
[EPR1157(2)] - Low endotoxin, Azide free
(ab224797)

This IHC data was generated using the same anti-CD80 antibody clone [EPR1157(2)] in a different buffer formulation (cat# **ab134120**).

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human tonsil tissue labelling CD80 with ab134120 at a dilution of 1/1000. Heat mediated antigen retrieval was performed using Trs/EDTA buffer, pH9 (ab93684). ImmunoHistoProbe one step HRP Polymer was used. Counter stained with hematoxylin.

The image shows cytoplasmic and membrane staining.



Western blot - Anti-CD80 antibody [EPR1157(2)] - Low endotoxin, Azide free (ab224797)

All lanes : Anti-CD80 antibody [EPR1157(2)] (<u>ab134120</u>) at 1/1000 dilution

Lane 1 : Wild-type Raji (Human Burkitt's lymphoma cell line) whole cell lysate

Lane 2 : CD80 knockout Raji (Human Burkitt's lymphoma cell line) whole cell lysate

Lane 3 : Ramos (Human Burkitt's lymphoma cell line) whole cell lysate

Lysates/proteins at 20 µg per lane.

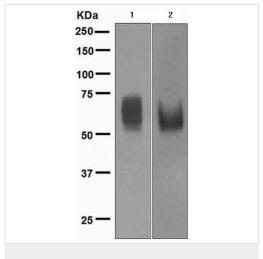
Performed under reducing conditions.

Predicted band size: 33 kDa **Observed band size:** 55 kDa

This data was developed using the same antibody clone in a different buffer formulation (ab134120).

Lanes 1 - 3: Merged signal (red and green). Green - <u>ab134120</u> observed at 55 kDa. Red - loading control <u>ab7291</u> (Mouse anti-Alpha Tubulin [DM1A]) observed at 55kDa.

ab134120 was shown to react with CD80 in wild-type Raji cells in western blot with loss of signal observed in CD80 knockout sample. Wild-type and CD80 knockout Raji cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% milk in TBS-T (0.1% Tween®) before incubation with ab134120 and ab7291 (Mouse anti-Alpha Tubulin [DM1A]) overnight at 4°C at a 1 in 1000 Dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-CD80 antibody [EPR1157(2)] - Low endotoxin, Azide free (ab224797)

All lanes : Anti-CD80 antibody [EPR1157(2)] (<u>ab134120</u>) at 1/1000 dilution (Unpurified)

Lane 1 : Raji cell lysate

Lane 2 : Ramos cell lysate

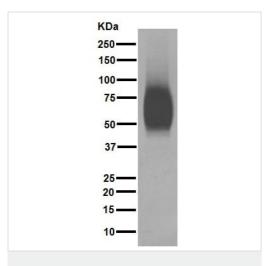
Lysates/proteins at 10 µg per lane.

Secondary

All lanes : Goat anti-rabbit HRP conjugated antibody at 1/2000 dilution

Predicted band size: 33 kDa **Observed band size:** 60 kDa

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab224797)



Western blot - Anti-CD80 antibody [EPR1157(2)] - Low endotoxin, Azide free (ab224797)

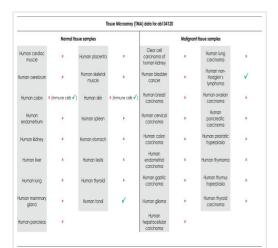
Anti-CD80 antibody [EPR1157(2)] (ab134120) at 1/2500 dilution (purified) + Raji cell lysate at 20 μg

Secondary

Peroxidase-conjugated goat anti-rabbit IgG (H+L) at 1/1000 dilution

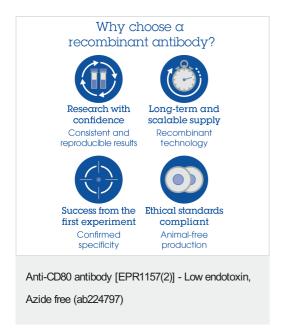
Predicted band size: 33 kDa **Observed band size:** 60 kDa

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab224797)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD80 antibody
[EPR1157(2)] - Low endotoxin, Azide free
(ab224797)

Tissue Microarrays stained for "Anti-CD80 antibody [EPR1157(2)]" using "ab134120" in immunohistochemical analysis. This table provides a detailed overview of positive (tick mark) and negative (cross mark) staining per sample type tested. The sections were pre-treated using Heat mediated antigen retrieval using Bond™ Epitope Retrieval Solution 2 (pH 9.0) for 20 minutes. The sections were incubated with ab134120 for 30 mins at room temperature followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101). The immunostaining was performed on a Leica Biosystems BOND® RX instrument.



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