abcam

Product datasheet

Anti-CD68 antibody [EPR23917-164] - BSA and Azide free ab283667



製品の概要

リコンピナント

RabMAb

**** 1 Abreviews 画像数 15

製品名 Anti-CD68 antibody [EPR23917-164] - BSA and Azide free

製品の詳細 Rabbit monoclonal [EPR23917-164] to CD68 - BSA and Azide free

由来種 Rabbit

特異性 Mouse unsuitable for IHC-Fr application

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

適用なし: №

種交差性 交差種: Mouse, Rat

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: Mouse spleen, RAW264.7, Mouse spleen, J774A.1, Rat liver and Rat spleen lysates. IHC-P:

Mouse spleen, Mouse colon, Mouse large B lymphoma, Rat spleen, Rat lung and Rat liver tissues.

IHC-Fr: and Rat liver, Rat spleen tissues. ICC/IF: J774A.1 cells. Flow Cyt: RAW 264.7 cell.

特記事項 ab283667 is the carrier-free version of ab283654.

> 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 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**.

製品の特性

製品の状態 Liquid

保存方法 Shipped at 4°C. Store at +4°C.

バッファー pH: 7.2

Constituent: 100% PBS

キャリア・フリー はい

精製度 Protein A purified

ポリ/モノ モノクローナル

クローン名 EPR23917-164

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ab283667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
IHC-P	****(1)	Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
ICC/IF		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Predicted molecular weight: 37 kDa.
IHC-Fr		Use at an assay dependent concentration. Mouse unsuitable for IHC-Fr application
Flow Cyt (Intra)		Use at an assay dependent concentration.

追加情報 Is unsuitable for IP.

ターゲット情報

機能

Could play a role in phagocytic activities of tissue macrophages, both in intracellular lysosomal metabolism and extracellular cell-cell and cell-pathogen interactions. Binds to tissue- and organ-specific lectins or selectins, allowing homing of macrophage subsets to particular sites. Rapid recirculation of CD68 from endosomes and lysosomes to the plasma membrane may allow macrophages to crawl over selectin-bearing substrates or other cells.

組織特異性 Highly expressed by blood monocytes and tissue macrophages. Also expressed in lymphocytes,

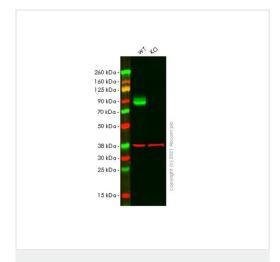
fibroblasts and endothelial cells. Expressed in many tumor cell lines which could allow them to attach to selectins on vascular endothelium, facilitating their dissemination to secondary sites.

配列類似性 Belongs to the LAMP family.

翻訳後修飾 N- and O-glycosylated.

細胞内局在 Cell membrane and Endosome membrane. Lysosome membrane.

画像



Western blot - Anti-CD68 antibody [EPR23917-164]

- BSA and Azide free (ab283667)

All lanes : Anti-CD68 antibody [EPR23917-164] (**ab283654**) at 1/1000 dilution

Lane 1 : Wild-type RAW264.7 (mouse Abelson murine leukemia

virus-induced tumor macrophage) whole cell lysate

Lane 2: Mouse CD68 knockout RAW 264.7 cell lysate

(ab280106)

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG H&L (IRDye® 800CW) (ab216773) and Goat Anti-Mouse lgG H&L (IRDye® 680RD)

(ab216776) at 1/10000 dilution

Predicted band size: 37 kDa **Observed band size:** 100 kDa

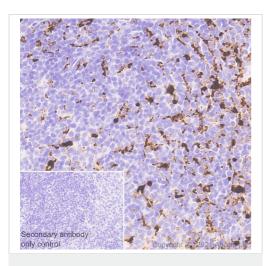
This data was developed using <u>ab283654</u>, the same antibody clone in a different buffer formulation.

Blocking and diluting buffer and concentration: Intercept® (TBS) Blocking Buffer diluted with an equal volume of 0.1% TBS.

Lanes 1-2: Merged signal (red and green). Green - <u>ab283654</u> observed at 100 kDa. Red - loading control <u>ab8245</u> observed at 36 kDa.

<u>ab283654</u> Anti-CD68 antibody [EPR23917-164] was shown to specifically react with CD68 in wild-type RAW264.7 cells. Loss of signal was observed when knockout cell line (knockout cell lysate - <u>ab280106</u>) was used. Wild-type and CD68 knockout samples were subjected to SDS-PAGE. <u>ab283654</u> and Anti-GAPDH antibody [6C5] - Loading Control (<u>ab8245</u>) were incubated at 4? overnight at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were

developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (ab216776) secondary antibodies at 1 in 10000 dilution for 1 hour at room temperature before imaging.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD68 antibody

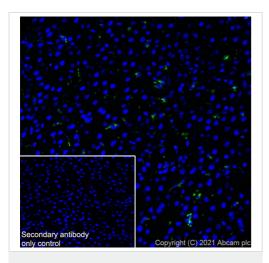
[EPR23917-164] - BSA and Azide free (ab283667)

This data was developed using <u>ab283654</u>, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded Mouse spleen tissue labelling CD68 with <u>ab283654</u> at 1/100 (4.66 ug/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining on mouse spleen. The section was incubated with <u>ab283654</u> for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins



Immunohistochemistry (Frozen sections) - Anti-CD68 antibody [EPR23917-164] - BSA and Azide free (ab283667)

This data was developed using <u>ab283654</u>, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of 4% PFA-fixed, 0.2% Triton X-100 permeabilized frozen Rat liver (fresh) tissue labeling CD68 with **ab283654** at 1/100 (4.66 ug/ml) dilution followed by **ab150081** Goat Anti-Rabbit lgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 (2 ug/ml) dilution (Green). Positive staining on Kupffer cells of rat liver is observed. The nuclear counterstain was DAPI (Blue).

Secondary antibody control: Secondary antibody is <u>ab150081</u> Goat Anti-Rabbit lgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 (2 ug/ml) dilution.



Western blot - Anti-CD68 antibody [EPR23917-164] - BSA and Azide free (ab283667)

All lanes : Anti-CD68 antibody [EPR23917-164] (**ab283654**) at 1/1000 dilution

Lane 1: Mouse spleen tissue lysate

Lane 2 : Mouse skeletal muscle tissue lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes: Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated

(ab97051) at 1/50000 dilution

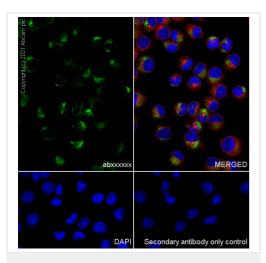
Predicted band size: 37 kDa **Observed band size:** 100 kDa

This data was developed using <u>ab283654</u>, the same antibody clone in a different buffer formulation.

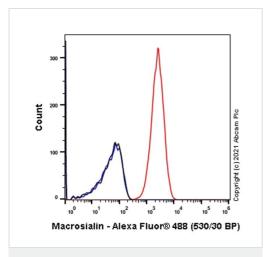
Blocking and diluting buffer and concentration: 5% NFDM/TBST

Negative control: mouse skeletal muscle (PMID: 28091823).

Exposure time: 37 seconds



Immunocytochemistry/ Immunofluorescence - Anti-CD68 antibody [EPR23917-164] - BSA and Azide free (ab283667)



Flow Cytometry - Anti-CD68 antibody [EPR23917-164] - BSA and Azide free (ab283667)

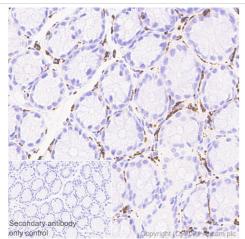
This data was developed using <u>ab283654</u>, the same antibody clone in a different buffer formulation.

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized J774A.1 cells labelling CD68 with ab283654 at 1/50 (9.32 ug/ml) dilution, followed by ab150081 Goat Anti-Rabbit lgG H&L (Alexa Fluor® 488) preadsorbed antibody at 1/1000 (2 ug/ml) dilution (Green). Confocal image showing cytoplasmic staining in J774A.1 cell line. ab195889 Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor® 594) was used to counterstain tubulin at 1/200 (2.5 ug/ml) dilution (Red). The Nuclear counterstain was DAPI (Blue).

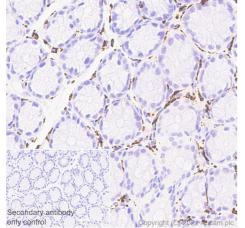
Secondary antibody only control: Secondary antibody is ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 (2 ug/ml) dilution.

This data was developed using <u>ab283654</u>, the same antibody clone in a different buffer formulation.

Flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized RAW 264.7 (mouse Abelson murine leukemia virus-induced tumor macrophage) cells labelling CD68 with ab283654 at 1/500 dilution (0.1ug) (Red) compared with a Rabbit monoclonal lgG (ab172730) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat F(ab')2 Anti-Rabbit lgG(DyLight® 488, ab98507) at 1/500 dilution was used as the secondary antibody.



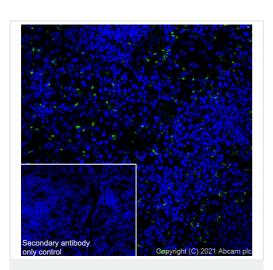
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD68 antibody [EPR23917-164] - BSA and Azide free (ab283667)



This data was developed using ab283654, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of 4% PFA-fixed, 0.2% Triton X-100 permeabilized frozen Rat spleen (fresh) tissue labeling CD68 with ab283654 at 1/100 (4.66 ug/ml) dilution followed by ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 (2 ug/ml) dilution (Green). Positive staining on macrophage of rat spleen is observed. The nuclear counterstain was DAPI (Blue).

Secondary antibody control: Secondary antibody is ab150081 Goat Anti-Rabbit lgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 (2 ug/ml) dilution.



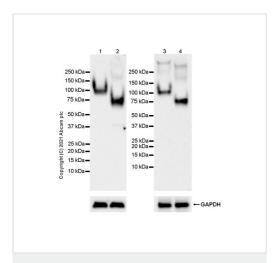
Immunohistochemistry (Frozen sections) - Anti-CD68 antibody [EPR23917-164] - BSA and Azide free (ab283667)

This data was developed using **ab283654**, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded Mouse colon tissue labelling CD68 with ab283654 at 1/100 (4.66 ug/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining on immune cells in mouse colon. The section was incubated with ab283654 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins



Western blot - Anti-CD68 antibody [EPR23917-164] - BSA and Azide free (ab283667)

All lanes : Anti-CD68 antibody [EPR23917-164] (**ab283654**) at 1/1000 dilution

Lane 1 : RAW264.7 (mouse Abelson murine leukemia virusinduced tumor macrophage), whole cell lysate

Lane 2: RAW264.7 treated with PNGase F whole cell lysate

Lane 3: Mouse spleen tissue lysate

Lane 4: Mouse spleen treated with PNGase F tissue lysate

Lysates/proteins at 15 µg per lane.

Secondary

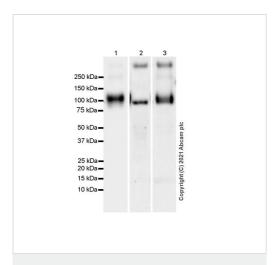
All lanes : Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated (ab97051) at 1/20000 dilution

Predicted band size: 37 kDa **Observed band size:** 100 kDa

This data was developed using <u>ab283654</u>, the same antibody clone in a different buffer formulation.

Blocking and diluting buffer and concentration: 5% NFDM/TBST Macrosialin (CD68) is a glycoprotein and can be de-glycosylated by PNGase F. The molecular mass observed is consistent with the literature (PMID: 7680921)

Exposure time: Lane 1-2: 7.75 seconds; Lane 3-4: 48 seconds



Western blot - Anti-CD68 antibody [EPR23917-164] - BSA and Azide free (ab283667)

All lanes : Anti-CD68 antibody [EPR23917-164] (**ab283654**) at 1/1000 dilution

Lane 1 : J774A.1 (mouse reticum cell sarcoma monocyte macrophage) whole cell lysate 20

Lane 2 : Rat liver tissue lysate

Lane 3 : Rat spleen tissue lysate

Lysates/proteins at 20 µg per lane.

Secondary

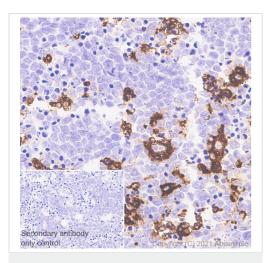
All lanes : Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated (ab97051) at 1/50000 dilution

Predicted band size: 37 kDa **Observed band size:** 100 kDa

This data was developed using <u>ab283654</u>, the same antibody clone in a different buffer formulation.

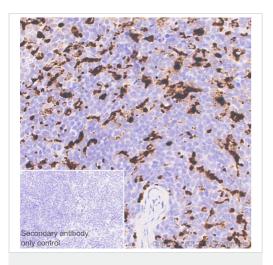
Blocking and diluting buffer and concentration: 5% NFDM/TBST

Exposure time: Lane 1: 7.75 seconds; Lane 2: 125 seconds; Lane 3: 92 seconds



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD68 antibody

[EPR23917-164] - BSA and Azide free (ab283667)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD68 antibody
[EPR23917-164] - BSA and Azide free (ab283667)

This data was developed using <u>ab283654</u>, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded Mouse large B cell lymphoma tissue labelling CD68 with <u>ab283654</u> at 1/100 (4.66 ug/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining on mouse large B cell lymphoma. The section was incubated with <u>ab283654</u> for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

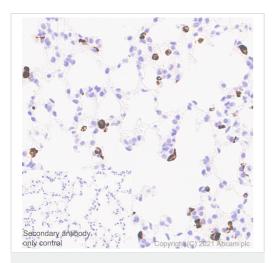
Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins

This data was developed using <u>ab283654</u>, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded Rat spleen tissue labelling CD68 with <u>ab283654</u> at 1/100 (4.66 ug/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining on rat spleen. The section was incubated with <u>ab283654</u> for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin.

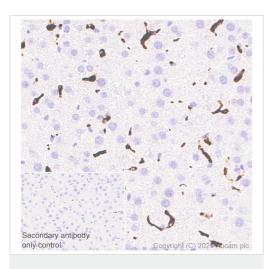
Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD68 antibody

[EPR23917-164] - BSA and Azide free (ab283667)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD68 antibody

[EPR23917-164] - BSA and Azide free (ab283667)

This data was developed using <u>ab283654</u>, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded Rat lung tissue labelling CD68 with <u>ab283654</u> at 1/100 (4.66 ug/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining on macrophages in rat lung. The section was incubated with <u>ab283654</u> for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

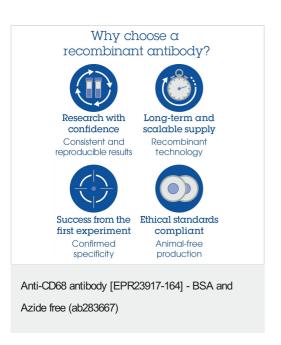
Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins

This data was developed using <u>ab283654</u>, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded Rat liver tissue labelling CD68 with <u>ab283654</u> at 1/100 (4.66 ug/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining on Kupffer cells in rat liver. The section was incubated with <u>ab283654</u> for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins



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