abcam

Product datasheet

Anti-CD68 antibody [EPR23917-164] ab283654





重组 RabMAb

★★★★★ 1 Abreviews 9 References 15 图像

概述

产品名称 Anti-CD68抗体[EPR23917-164]

描述 兔单克隆抗体[EPR23917-164] to CD68

宿主 Rabbit

特异性 Mouse unsuitable for IHC-Fr application

经测试应用 适用于: IHC-P, IHC-Fr, ICC/IF, Flow Cyt (Intra), WB

不适用于: №

种属反应性 与反应: Mouse, Rat

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

阳性对照 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.

常规说明 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 short term (1-2 weeks). Upon delivery aliquot. Store at -20°C.

Avoid freeze / thaw cycle.

存储溶液 pH: 7.2

Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

纯度 Protein A purified

克隆 单克隆

克隆编号 EPR23917-164

同种型 IgG

应用

The Abpromise guarantee Abpromise™承诺保证使用ab283654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
IHC-P	****(1)	1/100. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
IHC-Fr		1/100. Mouse unsuitable for IHC-Fr application
ICC/IF		1/50.
Flow Cyt (Intra)		1/500.
WB		1/1000. Predicted molecular weight: 37 kDa.

应用说明 Is unsuitable for IP.

靶标

功能 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 organspecific 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.

组织特异性 Highly expressed by blood monocytes and tissue macrophages. Also expressed in lymphocytes,

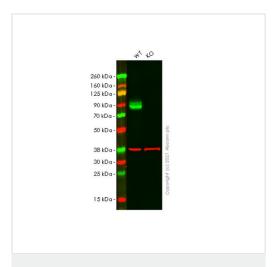
fibroblasts and endothelial cells. Expressed in many tumor cell lines which could allow them to attach to selectins on vascular endothelium, facilitating their dissemination to secondary sites.

序列相似性 Belongs to the LAMP family.

翻译后修饰 N- and O-glycosylated.

细胞定位 Cell membrane and Endosome membrane. Lysosome membrane.

图片



Western blot - Anti-CD68 antibody [EPR23917-164] (ab283654)

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

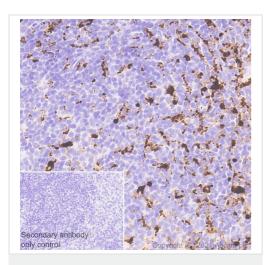
All lanes: Goat Anti-Rabbit IgG H&L (IRDye® 800CW)
(ab216773) and Goat Anti-Mouse IgG H&L (IRDye® 680RD)
(ab216776) at 1/10000 dilution

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

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 - ab283654 observed at 100 kDa. Red - loading control ab8245 observed at 36 kDa.

ab283654 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 - ab280106) was used. Wild-type and CD68 knockout samples were subjected to SDS-PAGE. ab283654 and Anti-GAPDH antibody [6C5] - Loading Control (ab8245) were incubated at 4? overnight at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (ab216773) and Goat anti-Mouse lgG H&L (IRDye® 680RD) preadsorbed (ab216776) secondary antibodies at 1 in 10000 dilution for 1 hour at room temperature before imaging.

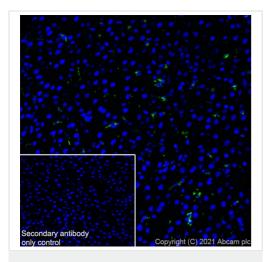


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD68 antibody
[EPR23917-164] (ab283654)

Immunohistochemical analysis of paraffin-embedded Mouse spleen 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 mouse spleen. 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).

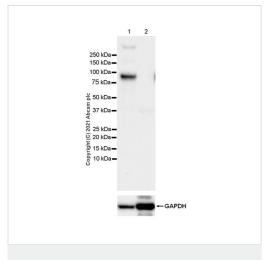
Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins



Immunohistochemistry (Frozen sections) - Anti-CD68 antibody [EPR23917-164] (ab283654)

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] (ab283654)

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 IgG, (H+L), Peroxidase conjugated

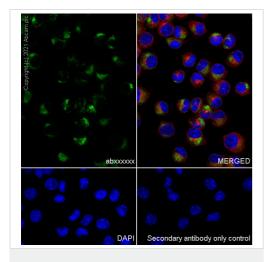
(ab97051) at 1/50000 dilution

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

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

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

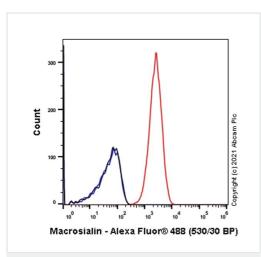
Exposure time: 37 seconds



Immunocytochemistry/ Immunofluorescence - Anti-CD68 antibody [EPR23917-164] (ab283654)

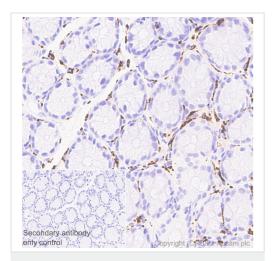
Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized J774A.1 cells labelling CD68 with ab283654 at 1/50 (9.32 ug/ml) dilution, followed by <u>ab150081</u> Goat Anti-Rabbit lgG H&L (Alexa Fluor® 488) preadsorbed antibody at 1/1000 (2 ug/ml) dilution (Green). Confocal image showing cytoplasmic staining in J774A.1 cell line. <u>ab195889</u> 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 <u>ab150081</u> Goat Anti-Rabbit lgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 (2 ug/ml) dilution.



Flow Cytometry (Intracellular) - Anti-CD68 antibody [EPR23917-164] (ab283654)

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 IgG (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 IgG(DyLight® 488, ab98507) at 1/500 dilution was used as the secondary antibody.

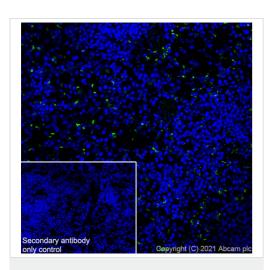


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD68 antibody
[EPR23917-164] (ab283654)

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).

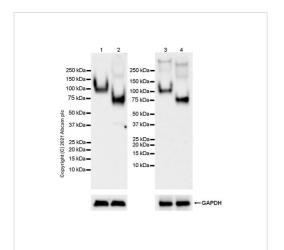
Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins



Immunohistochemistry (Frozen sections) - Anti-CD68 antibody [EPR23917-164] (ab283654)

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 lgG 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 <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] (ab283654)

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

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] (ab283654)

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.

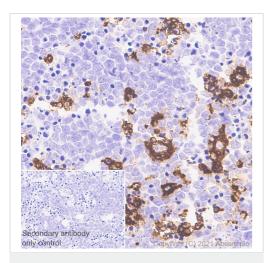
Secondary

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

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

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

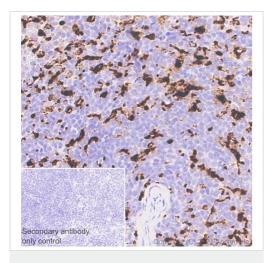


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD68 antibody
[EPR23917-164] (ab283654)

Immunohistochemical analysis of paraffin-embedded Mouse large B cell lymphoma 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 mouse large B cell lymphoma. 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

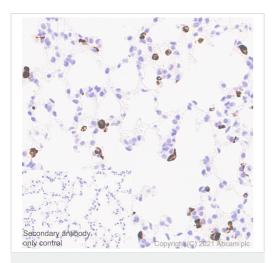


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD68 antibody
[EPR23917-164] (ab283654)

Immunohistochemical analysis of paraffin-embedded Rat spleen 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 rat spleen. 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

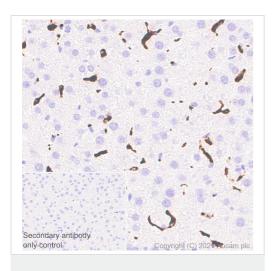


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD68 antibody
[EPR23917-164] (ab283654)

Immunohistochemical analysis of paraffin-embedded Rat lung 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 macrophages in rat lung. 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

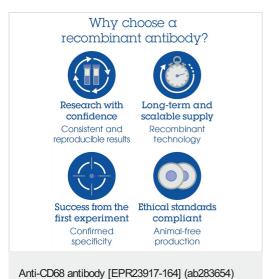


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD68 antibody
[EPR23917-164] (ab283654)

Immunohistochemical analysis of paraffin-embedded Rat liver 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 Kupffer cells in rat liver. 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



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