abcam

## Product datasheet

# Anti-F4/80 antibody [CI:A3-1] ab6640

59 Abreviews 390 References 8 Images

### Overview

| | |
|---|---|
| **Product name** | Anti-F4/80 antibody [CI:A3-1] |
| **Description** | Rat monoclonal [CI:A3-1] to F4/80 |
| **Host species** | Rat |
| **Specificity** | This antibody recognises the mouse F4/80 antigen, a 160kD glycoprotein expressed by murine macrophages. Abcam does not recommend this product for IHC-P. |
| **Tested applications** | **Suitable for:** WB, IP, RIA, IHC-Fr, Flow Cyt, ICC, IHC-FoFr, ICC/IF, IHC-R |
| **Species reactivity** | **Reacts with:** Mouse |
| **Immunogen** | Thioglycollate stimulated peritoneal macrophages from C57/BL mice |
| **Positive control** | WB: Mouse spleen and macrophages lysate. ICC/IF: Raw264.7 cells; Mouse bone marrow cells. IHC-Fr: Mouse spleen and intestine tissue. Flow Cyt: J774 cells and peritoneal macrophages. |
| **General notes** | **Please note we cannot guarantee IHC-P, however should you use this application for this product then please use a specific protocol which can be found in the protocols section of our datasheet here.** |

### Properties

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|---|---|
| **Form** | Liquid |
| **Storage instructions** | Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C or -80°C. Avoid freeze / thaw cycle. |
| **Storage buffer** | Preservative: 0.09% Sodium azide<br>Constituent: PBS |
| **Purity** | Protein G purified |
| **Purification notes** | Purified IgG prepared by affinity chromatography on Protein G from tissue culture supernatant. |
| **Clonality** | Monoclonal |
| **Clone number** | CI:A3-1 |
| **Myeloma** | NS1 |
| **Isotype** | IgG2b |

### Applications

Our Abpromise guarantee covers the use of **ab6640** in the following tested applications.

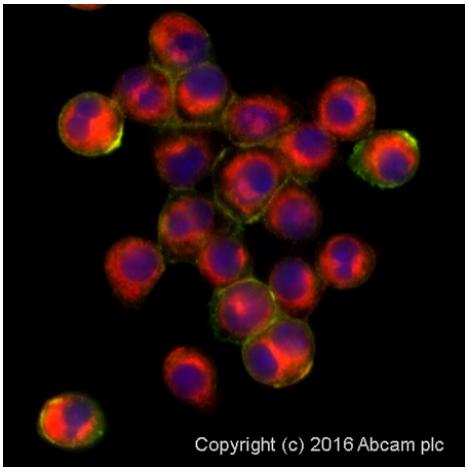
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

| Application | Abreviews | Notes |
|---|---|---|
| WB | ★★★★☆ | 1/1000. Use under non reducing condition. Detects a band of approximately 160 kDa (predicted molecular weight: 102 kDa).<br>(predicted molecular weight of precursor protein: 102 kDa; protein is heavily glycosylated). Block in 5% milk for 1 hour. |
| IP | | Use at an assay dependent concentration. |
| RIA | | Use at an assay dependent concentration. |
| IHC-Fr | ★★★★★ | Use a concentration of 10 µg/ml. |
| Flow Cyt | | 1/10 - 1/50.<br>ab18536 - Rat monoclonal IgG2b, is suitable for use as an isotype control with this antibody. |
| ICC | ★★★★★ | Use at an assay dependent concentration. |
| IHC-FoFr | ★★★★☆ | Use a concentration of 10 µg/ml. |
| ICC/IF | ★★★★★ | Use a concentration of 5 - 10 µg/ml.<br>We recommend ab150165 Goat Anti-Rat IgG H&L (Alexa Fluor® 488) secondary antibody. |
| IHC-R | | Use a concentration of 10 µg/ml. |

## Target

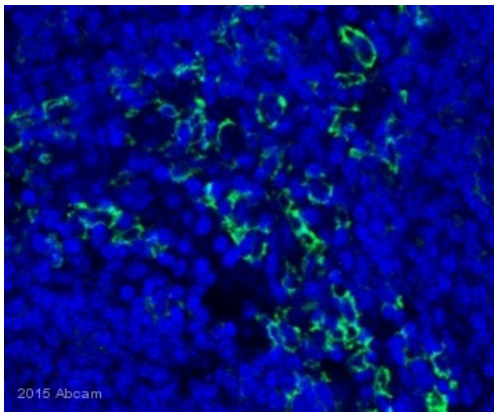
| | |
|---|---|
| **Function** | Orphan receptor involved in cell adhesion and probably in cell-cell interactions specifically involving cells of the immune system. May play a role in regulatory T-cells (Treg) development. |
| **Tissue specificity** | Expression is restricted to eosinophils. |
| **Sequence similarities** | Belongs to the G-protein coupled receptor 2 family. Adhesion G-protein coupled receptor (ADGR) subfamily.<br>Contains 6 EGF-like domains.<br>Contains 1 GPS domain. |
| **Cellular localization** | Cell membrane. |

## Images




Immunocytochemistry/ Immunofluorescence - Anti-F4/80 antibody [Cl:A3-1] (ab6640)

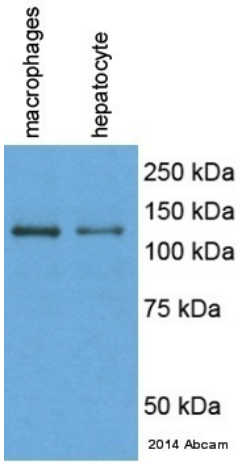
ab6640 stained Raw 264.7 cells. The cells were 100% methanol fixed for 5 minutes at room temperatureand then incubated in 1%BSA / 10% normal Goat serum / 0.3M glycine in 0.1% PBS-Tween for 1hour at room temperature to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody (ab6640 at 5µg/ml) overnight at +4°C. The secondary antibody (pseudo-colored green) was ab150165 Goat Anti-Rat IgG H&L (Alexa Fluor® 488) preadsorbed used at a 1/1000 dilution for 1 hour at room temperature. Alexa Fluor® 594 WGA was used to label plasma membranes (pseudo-colored red) at a 1/200 dilution for 1hour at room temperature. DAPI was used to stain the cell nuclei (pseudo-colored blue) at a concentration of 1.43µM for 1hour at room temperature.




Immunohistochemistry (Frozen sections) - Anti-F4/80 antibody [Cl:A3-1] (ab6640)

This image is courtesy of an anonymous Abreview

ab6640 staining F4/80 in mouse spleen tissue sections by Immunohistochemistry (PFA perfusion fixed frozen sections). Tissue samples were fixed by perfusion with paraformaldehyde and blocked with 5% serum for 45 minutes at 21°C. The sample was incubated with primary antibody (1/100 in PBS + 5% FBS) at 4°C for 16 hours. An Alexa Fluor® 488-conjugated goat anti-rat IgG (H+L) polyclonal (1/200) was used as the secondary antibody.




Western blot - Anti-F4/80 antibody [CI:A3-1] (ab6640)

This image is courtesy of an anonymous Abreview

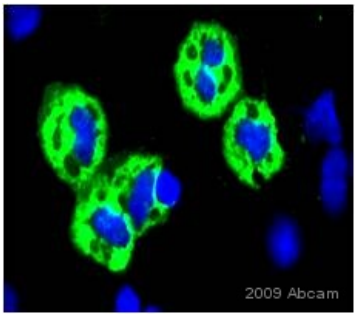
**All lanes :** Anti-F4/80 antibody [CI:A3-1] (ab6640) at 1/1000 dilution

**Lane 1 :** Mouse macrophage lysates
**Lane 2 :** Mouse hepatocyte lysates

Lysates/proteins at 20 µg per lane.

**Secondary**
**All lanes :** HRP labelled donkey anti-rat at 1/5000 dilution

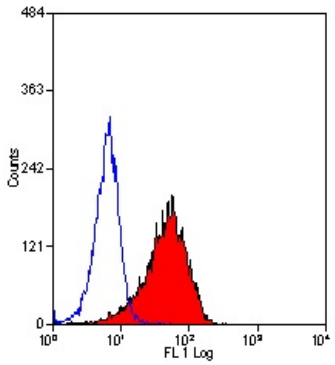
**Predicted band size:** 102 kDa
**Observed band size:** 130 kDa
why is the actual band size different from the predicted?




Immunocytochemistry/ Immunofluorescence - Anti-F4/80 antibody [CI:A3-1] (ab6640)

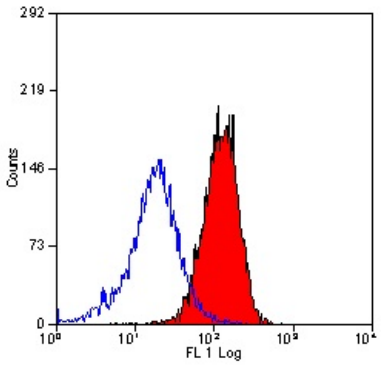
This image is courtesy of an anonymous Abreview

ab6640 staining F4/80 in mouse bone marrow cells by immunocytochemistry/ immunofluorescence. Cells were formaldehyde fixed and permeabilized in 0.2% Triton X-100 prior to blocking in 2% BSA for 30 minutes at 20°C. The primary antibody was diluted 1/200 and incubated with the sample for 9 hour at 4°C. Alexa fluor® 488 goat polyclonal to rat Ig, diluted 1/200, was used as the secondary antibody.



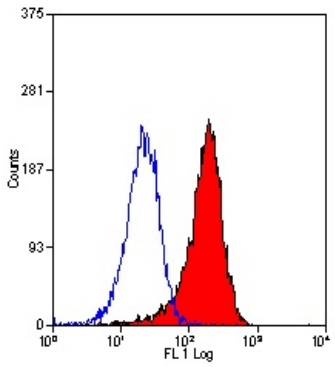

Flow Cytometry - Anti-F4/80 antibody [Cl:A3-1] (ab6640)

Flow cytometry analysis of J774 cells labelling F4/80 (red) with ab6640 at a dilution of 1/10 followed by streptavidin FITC secondary at 1/100 dilution. The blue line shows J774 cells stained with Rat anti mouse isotype control.



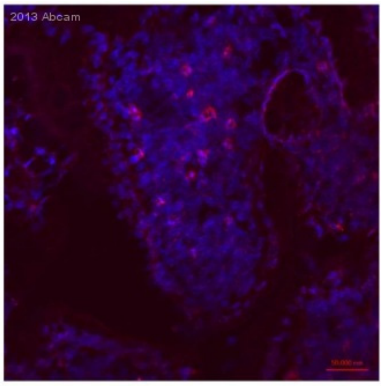

Flow Cytometry - Anti-F4/80 antibody [Cl:A3-1] (ab6640)

Flow cytometry analysis of peritoneal macrophages labelling F4/80 (red) with ab6640 at a dilution of 1/10. The blue line shows J774 cells stained with Rat anti mouse isotype control.




Flow Cytometry - Anti-F4/80 antibody [Cl:A3-1] (ab6640)

Flow cytometry analysis of J774 cells labelling F4/80 (red) with ab6640 at a dilution of 1/50 followed by goat anti rat IgG FITC secondary antibody at 1/100 dilution. The blue line shows J774 cells stained with Rat anti mouse isotype control.




Immunohistochemistry (Frozen sections) - Anti-F4/80 antibody [Cl:A3-1] (ab6640)

This image is courtesy of an anonymous Abreview

ab6640 staining F4/80 (red) in Mouse intestine tissue sections by Immunohistochemistry (IHC-Fr - frozen sections). Tissue was fixed with acetone and blocked with 10% serum for 1 hour at 21°C. Samples were incubated with primary antibody (1/10µg/ml in PBS + 10% serum) for 16 hours at 4°C. An Alexa Fluor® 555-conjugated Donkey anti-rat IgG polyclonal (1/1000) was used as the secondary antibody. Blue - nuclei.



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