# Anti-Iba1 antibody ab5076

**Overview**

<table>
<thead>
<tr>
<th><strong>Product name</strong></th>
<th>Anti-Iba1 antibody</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Description</strong></td>
<td>Goat polyclonal to Iba1</td>
</tr>
<tr>
<td><strong>Host species</strong></td>
<td>Goat</td>
</tr>
<tr>
<td><strong>Specificity</strong></td>
<td>ab5076 is expected to recognize the isoforms represented by NP_001614 and NP_116573 but not NP_004838.</td>
</tr>
</tbody>
</table>

**Tested applications**

- Suitable for: Electron Microscopy, IHC-P, WB, IHC-FrFl, ICC, ICC/IF
- Unsuitable for: IHC-FoFr

**Species reactivity**

- Reacts with: Rat, Rabbit, Guinea pig, Cow, Dog, Human, Common marmoset
- Predicted to work with: Macaque monkey

**Immunogen**

- Synthetic peptide corresponding to Human Iba1 aa 135-147 (C terminal). Accession Number(s): NP_116573.1; NP_001614.3
- Sequence: C-TGPPAKKAISELP
- Database link: P55008 (Peptide available as ab23067)

**Positive control**

Recombinant Human Iba1 protein (ab117478) can be used as a positive control in WB. WB: Human lymph node; Rat brain lysate.

**General notes**

The Iba1 antibody (ab5076) is commonly used as a marker of microglia activation in staining and immunohistochemistry, given that ionized calcium binding adaptor molecule 1 (Iba1) is a microglia/macrophage-specific calcium-binding protein with actin-bundling activity that participates in membrane ruffling and phagocytosis in activated microglia.

**Properties**

<table>
<thead>
<tr>
<th><strong>Form</strong></th>
<th>Liquid</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Storage instructions</strong></td>
<td>Shipped at 4°C. Upon delivery aliquot and store at -20°C. Avoid freeze / thaw cycles.</td>
</tr>
</tbody>
</table>
| **Storage buffer** | pH: 7.30  
Preservative: 0.02% Sodium azide |
Constituents: Tris buffered saline, 0.5% BSA

Purity
Immunogen affinity purified

Purification notes
Purified from goat serum by ammonium sulphate precipitation followed by antigen affinity chromatography using the immunizing peptide.

Clonality
Polyclonal

Isotype
IgG

Applications

Our Abpromise guarantee covers the use of ab5076 in the following tested applications.

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

<table>
<thead>
<tr>
<th>Application</th>
<th>Abreviews</th>
<th>Notes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Electron Microscopy</td>
<td></td>
<td>Use at an assay dependent concentration. PubMed: 26358247</td>
</tr>
<tr>
<td>IHC-P</td>
<td>★★★★★</td>
<td>Use a concentration of 10 µg/ml. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. Heat induced antigen retrieval with Tris-EDTA pH 9</td>
</tr>
<tr>
<td>WB</td>
<td>★★★★★</td>
<td>Use a concentration of 1 - 3 µg/ml. Detects a band of approximately 16-17 kDa (predicted molecular weight: 17 kDa). Can be blocked with Human Iba1 peptide (ab23067). A 1 hour primary incubation is recommended for this product. We received several excellent Abreviews on this antibody working with mouse, however, mouse is not a batch tested species and we cannot guarantee that this antibody will work on mouse. Some of our customers have observed high background in mouse samples. We recommend blocking in milk (3% milk in TBST) for 1 hour at room temperature. Blocking with BSA gives high background.</td>
</tr>
<tr>
<td>IHC-FrF</td>
<td>★★★★★</td>
<td>Use at an assay dependent concentration.</td>
</tr>
<tr>
<td>ICC</td>
<td>★★★★★</td>
<td>Use at an assay dependent concentration.</td>
</tr>
<tr>
<td>ICC/IF</td>
<td>★★★★★</td>
<td>Use at an assay dependent concentration.</td>
</tr>
</tbody>
</table>

Application notes
Is unsuitable for IHC-FoFr.

Target

Function

Tissue specificity
Detected in T-lymphocytes and peripheral blood mononuclear cells.

Sequence similarities
Contains 2 EF-hand domains.

Post-translational modifications
Phosphorylated on serine residues.
**Cellular localization**

Cytoplasm > cytoskeleton. Cell projection > ruffle membrane. Associated with the actin cytoskeleton at membrane ruffles and at sites of phagocytosis.

**Images**

IHC-P image of Iba1 staining on cow kidney sections using ab5076 (1:2000). The sections were deparaffinized and subjected to heat mediated antigen retrieval using citric acid. The sections were blocked using 1% BSA for 10 mins at 21°C. ab5076 was diluted 1:2000 and incubated with the sections for 2 hours at 21°C. The secondary antibody used was Rabbit polyclonal to anti sheep/goat IgG conjugated to biotin (1:200).

ICC/IF image Iba1 staining on rat 33B (N-Ethyl-N-nitrosourea-induced rat neural tumour cell line) whole cell using ab5076 (1/500). The cells were fixed in Paraformaldehyde and blocked with 10% serum for 20 minutes at 24°C. ab5076 was diluted 1/500 in PBS and incubated with the cells for 16 hours at 4°C. The secondary antibody used was Rabbit Anti Goat Alexa Flour® 555-conjugated (1/1000).
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Iba1 antibody (ab5076)
This image is courtesy of an abreview submitted by Carl Hobbs (King's College London, United Kingdom)

IHC-P image of Iba1 staining on cat kidney sections using ab5076 (1:1000). The sections were deparaffinized and subjected to heat mediated antigen retrieval using citric acid. The sections were blocked using 1% BSA for 10 mins at 21°C. ab5076 was diluted 1:1000 and incubated with the sections for 2 hours at 21°C. The secondary antibody used was Rabbit polyclonal to anti sheep/goat IgG conjugated to biotin (1:200)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Iba1 antibody (ab5076)
This image is courtesy of an abreview submitted by Carl Hobbs (King's College London, United Kingdom)

IHC-P image of Iba1 staining on marmoset bladder sections using ab5076 (1:1000). The sections were deparaffinized and subjected to heat mediated antigen retrieval using citric acid. The sections were blocked using 1% BSA for 10 mins at 21°C. ab5076 was diluted 1:1000 and incubated with the sections for 2 hours at 21°C. The secondary antibody used was Rabbit polyclonal to anti goat IgG conjugated to biotin (1:200)

Immunohistochemical analysis of formalin-fixed, paraffin-embedded human lateral prefrontal cortex tissue labeling iba1 labeled microglia (labeled with black arrows) within the cortex using ab5076 at 1/1000 dilution. (Cropped image).

Sections were rinsed in 0.01 m PBS, pH 7.4, followed by 10% serum matching the species of the secondary antibody, 5% bovine serum albumin, and 0.1% Triton X-100 in 0.01 m PBS blocking solution for 1 h and incubated for 2 days at 4 °C in primary antibody. The sections were rinsed in PBS and incubated overnight at 4 °C with a horse anti-Goat IgG (Biotinylated) secondary antibody.
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Iba1 antibody (ab5076)

This image is a courtesy of Omer Butt

ab5076 staining Iba1 in guinea pig brain tissue section by Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections). Tissue underwent formaldehyde fixation before heat mediated antigen retrieval in sodium citrate and then blocking with 3% serum was performed for 30 minutes at RT. The primary antibody was used at dilution at 1/200 and incubated with sample at 2% blocking serum for 18 hours at 4°C. A Biotin conjugated horse polyclonal to goat IgG was used undiluted as secondary antibody.

AIF1 expression in an inflammatory foci in a skin biopsy. Paraffin embedded sections were deparaffinized and boiled in 10mM citrate buffer for 30 min in a microwave to expose the AIF1 antigen, and then blocked with 5% donkey serum for 20 min. Slides were incubated for 40 min with goat-anti-AIF1 antibody (1:100) and rinsed in three changes of PBS for 1 min each. A secondary antibody (donkey-anti-goat-FITC conjugated IgG; 1:50) was then applied to the slide for 40 min. Sections were washed again to remove the unbound antibody. The slides were counterstained with DaPI and viewed with a Nikon epi-fluorescent microscope. AIF1 positive cells appear green and the nuclei are stained blue. Numerous cells expressing the AIF1 protein are located around a vessel. (400X). Picture from Review by Carol Artlett submitted 30 July 2004.
**Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Iba1 antibody (ab5076)**

This image is courtesy of an abreview submitted by Carl Hobbs (King's College London, United Kingdom)

IHC-P image of Iba1 staining on rat brain sections using ab5076 (1:1000). The sections were deparaffinized and subjected to heat mediated antigen retrieval using citric acid. The sections were blocked using 1% BSA for 10 mins at 21°C. ab5076 was diluted 1:1000 and incubated with the sections for 2 hours at 21°C. The secondary antibody used was Rabbit polyclonal to anti sheep/goat IgG conjugated to biotin (1:200)

Free floating sections of rat striatum/microglia were stained for Iba1 with ab5076 at 1/2000 dilution in immunohistochemical analysis. Rabbit Anti-Goat IgG Biotin was used as the secondary antibody at 1/200 dilution.

**Western blot - Anti-Iba1 antibody (ab5076)**

All lanes: Anti-Iba1 antibody (ab5076) at 1 µg/ml

Lane 1: Human lymph node tissue lysate in RIPA buffer
Lane 2: Rat brain tissue lysate in RIPA buffer

Lysates/proteins at 35 µg per lane.

Developed using the ECL technique.

**Predicted band size**: 17 kDa

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