Product datasheet

Anti-YAP1 antibody [EP1674Y] ab52771

RabMAb

Overview

<table>
<thead>
<tr>
<th><strong>Product name</strong></th>
<th>Anti-YAP1 antibody [EP1674Y]</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Description</strong></td>
<td>Rabbit monoclonal [EP1674Y] to YAP1</td>
</tr>
<tr>
<td><strong>Host species</strong></td>
<td>Rabbit</td>
</tr>
<tr>
<td><strong>Tested applications</strong></td>
<td>Suitable for: WB, IP, Flow Cyt, IHC-P, ICC/IF</td>
</tr>
<tr>
<td><strong>Species reactivity</strong></td>
<td>Reacts with: Human</td>
</tr>
<tr>
<td><strong>Immunogen</strong></td>
<td>Synthetic peptide corresponding to Human YAP1 aa 350-450 (C terminal). (Peptide available as ab173007)</td>
</tr>
</tbody>
</table>

**Positive control**

- Purchase matching WB positive control: Recombinant Human YAP1 protein

**General notes**

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

Abcam recommended secondaries - Goat Anti-Rabbit HRP (ab205718) and Goat Anti-Rabbit Alexa Fluor® 488 (ab150077).

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMab® patents.

**We are constantly working hard to ensure we provide our customers with best in class antibodies. As a result of this work we are pleased to now offer this antibody in purified format. We are in the process of updating our datasheets. The purified format is designated 'PUR' on our product labels. If you have any questions regarding this update, please contact our Scientific Support team.**

This product is a recombinant rabbit monoclonal antibody.

Properties

**Form**

Liquid
Storage instructions


Storage buffer

pH: 7.20
Preservative: 0.01% Sodium azide
Constituents: PBS, 40% Glycerol, 0.5% BSA

Purity

Protein A purified

Clonality

Monoclonal

Clone number

EP1674Y

Isotype

IgG

Applications

Our Abpromise guarantee covers the use of ab52771 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>WB</td>
<td>★★★★★☆☆</td>
<td>1/5000. Detects a band of approximately 70 kDa (predicted molecular weight: 65 kDa).</td>
</tr>
<tr>
<td>IP</td>
<td>★★★★★☆☆</td>
<td>1/20. For unpurified use at 1:70.</td>
</tr>
<tr>
<td>Flow Cyt</td>
<td>★★★★★☆☆</td>
<td>1/20. ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody. For unpurified use at 1:50.</td>
</tr>
<tr>
<td>IHC-P</td>
<td>★★★★★☆☆</td>
<td>1/50.</td>
</tr>
<tr>
<td>ICC/IF</td>
<td>★★★★★☆☆</td>
<td>1/100 - 1/500.</td>
</tr>
</tbody>
</table>

Target

Function

Transcriptional regulator which can act both as a coactivator and a corepressor and is the critical downstream regulatory target in the Hippo signaling pathway that plays a pivotal role in organ size control and tumor suppression by restricting proliferation and promoting apoptosis. The core of this pathway is composed of a kinase cascade wherein MST1/MST2, in complex with its regulatory protein SAV1, phosphorylates and activates LATS1/2 in complex with its regulatory protein MOB1, which in turn phosphorylates and inactivates YAP1 oncoprotein and WWTR1/TAZ. Plays a key role to control cell proliferation in response to cell contact. Phosphorylation of YAP1 by LATS1/2 inhibits its translocation into the nucleus to regulate cellular genes important for cell proliferation, cell death, and cell migration. The presence of TEAD transcription factors are required for it to stimulate gene expression, cell growth, anchorage-independent growth, and epithelial mesenchymal transition (EMT) induction. Isoform 2 and isoform 3 can activate the C-terminal fragment (CTF) of ERBB4 (isoform 3).

Tissue specificity

Increased expression seen in some liver and prostate cancers. Isoforms lacking the transactivation domain found in striatal neurons of patients with Huntington disease (at protein
Sequence similarities
Belongs to the YORKIE family.
Contains 2 WW domains.

Post-translational modifications
Phosphorylated by LATS1 and LATS2; leading to cytoplasmic translocation and inactivation.
Phosphorylated by ABL1; leading to YAP1 stabilization, enhanced interaction with TP73 and recruitment onto proapoptotic genes; in response to DNA damage.

Cellular localization
Cytoplasm. Nucleus. Both phosphorylation and cell density can regulate its subcellular localization.
Phosphorylation sequesters it in the cytoplasm by inhibiting its translocation into the nucleus. At low density, predominantly nuclear and is translocated to the cytoplasm at high density.

Immunofluorescence revealed that YAP1 expression was significantly lower in iVSD hearts compared to control hearts.

YAP1 (green), and DAPI (blue) staining are shown; Scale bar = 25 μm.

Cryosections of human heart tissue were blocked using 10% FBS for 30 min, and then incubated with anti YAP1 (ab52771, 1:200) at room temperature for 2 h. The slides were then incubated with the following secondary antibody, Alexa Fluor® 488-conjugated anti-rabbit (Abcam, ab150073; 1:1,000 dilution). Cell nuclei were stained with DAPI.

Western blot - Anti-YAP1 antibody [EP1674Y] (ab52771) at 1/5000 dilution (purified) + HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysates at 15 μg

Secondary
Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

Predicted band size: 65 kDa

Blocking/Diluting buffer: 5% NFDM/TBST
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human thyroid carcinoma tissue sections labeling YAP1 with purified ab52771 at 1:50 dilution (1.78 µg/ml).

Heat mediated antigen retrieval was performed using ab93684 (Tris/EDTA buffer, pH 9.0). Tissue was counterstained with hematoxylin. ImmunoHistoProbe one step HRP Polymer (ready to use) was used as the secondary antibody.

PBS instead of the primary antibody was used as the negative control.

Immunocytochemistry/Immunofluorescence analysis of MCF-7 (Human breast adenocarcinoma cell line) cells labeling YAP1 with purified ab52771 at 1/500.

Cells were fixed with 4% paraformaldehyde and permeabilized by 0.1% Triton X-100. An Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/1000) was used as the secondary antibody (ab150077).

Nuclei counterstained with DAPI (blue).
Immunoprecipitation - Anti-YAP1 antibody (ab52771)

Lane 1: HeLa whole cell lysate 10 μg input.
Lane 2: ab52771 in HeLa whole cell lysate
Lane 3: Rabbit monoclonal IgG (ab172730) instead of ab52771 in HeLa whole cell lysate.

For western blotting, VeriBlot for IP secondary antibody (HRP) (ab131366) was used as the secondary antibody at 1:1000 dilution.

Blocking/Diluting buffer: 5% NFDM/TBST.

Flow Cytometry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling YAP1 with purified ab52771 at 1:20 dilution (10 µg/ml) (red).

Cells were fixed with 4% paraformaldehyde. A Goat anti rabbit IgG (Alexa Fluor® 488) secondary antibody was used at 1:2000 dilution. Isotype control - Rabbit monoclonal IgG (Black). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue).

Anti-YAP1 antibody [EP1674Y] (ab52771) at 1/50000 dilution (purified) + Human thyroid cancer lysates at 15 µg

Secondary
Rabbit monoclonal [EP1674Y] to YAP1 (ab52771) at 1/2000 dilution (Goat Anti-Rabbit IgG (HRP) with minimal cross-reactivity with human IgG)

Predicted band size: 65 kDa

Blocking/Dilution buffer: 5% NFDM/TBST
Immunohistochemical staining of YAP1 in paraffin embedded human breast carcinoma tissue using unpurified ab52771 at a 1/100 dilution.

Anti-YAP1 antibody [EP1674Y] (ab52771) at 1/50000 dilution (unpurified) + HeLa (Human epithelial cell line from cervix adenocarcinoma) cell lysate at 10 µg

**Secondary**
Goat anti-rabbit HRP at 1/2000 dilution

**Predicted band size:** 65 kDa
**Observed band size:** 65 kDa

Overlay histogram showing HeLa (Human epithelial cell line from cervix adenocarcinoma) cells stained with unpurified ab52771 (red line).

The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab52771, 1/50 dilution) for 30 min at 22°C. The secondary antibody used was DyLight® 488 goat anti-rabbit IgG (H+L) (ab96899) at 1/500 dilution for 30 min at 22°C.

Isotype control antibody (black line) was rabbit IgG (monoclonal) (1 µg/1x10^6 cells) used under the same conditions.

Acquisition of >5,000 events was performed.
All lanes are unpurified ab52771 at a 1/1000 dilution plus whole cell lysate prepared from U-87 MG (Human glioblastoma-astrocytoma epithelial cell line) cells, treated with DMSO or 1 μM and 10 μM LY294002, 50 μg positive control loaded.

Primary antibody was incubated for 16 hours at 4°C.

Blocking step was performed using 5% milk for 1 hour at 23°C.

Secondary used was a goat polyclonal conjugated to HRP, at a 1/10,000 dilution.

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