Anti-Integrin beta 1 antibody [EP1041Y] ab52971

Overview

Product name: Anti-Integrin beta 1 antibody [EP1041Y]
Description: Rabbit monoclonal [EP1041Y] to Integrin beta 1
Host species: Rabbit
Tested applications: Suitable for: WB, IHC-Fr, IHC-P
Unsuitable for: Flow Cyt or ICC/IF
Species reactivity: Reacts with: Human
Immunogen: Synthetic peptide within Human Integrin beta 1 aa 650-750. The exact sequence is proprietary.
Database link: P05556
General notes: 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: Shipped at 4°C. Upon delivery aliquot and store at -20°C. Avoid freeze / thaw cycles.
Storage buffer: pH: 7.20
Preservative: 0.01% Sodium azide
Constituents: 59% PBS, 40% Glycerol, 0.05% BSA
Purity: Protein A purified
Clonality: Monoclonal
Clone number: EP1041Y
Isotype  
IgG

Applications

Our Abpromise guarantee covers the use of ab52971 in the following tested applications.

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

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<tr>
<th>Application</th>
<th>Abreviews</th>
<th>Notes</th>
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<tbody>
<tr>
<td>WB</td>
<td>⭐⭐⭐⭐</td>
<td>1/1000 - 1/100000. Detects a band of approximately 140-150 kDa (predicted molecular weight: 88 kDa). For unpurified, use 1/500.</td>
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<tr>
<td>IHC-Fr</td>
<td>⭐⭐⭐⭐</td>
<td>Use at an assay dependent concentration.</td>
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<tr>
<td>IHC-P</td>
<td>⭐⭐⭐⭐</td>
<td>1/50. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. See IHC antigen retrieval protocols. Use of an HRP/AP polymerized secondary antibody is recommended. For unpurified, use 1/250 - 1/500.</td>
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</tbody>
</table>

Application notes  
Is unsuitable for Flow Cyt or ICC/IF.

Target

Function  
Integrins alpha-1/beta-1, alpha-2/beta-1, alpha-10/beta-1 and alpha-11/beta-1 are receptors for collagen. Integrins alpha-1/beta-1 and alpha-2/beta-2 recognize the proline-hydroxylated sequence G-F-P-G-E-R in collagen. Integrins alpha-2/beta-1, alpha-3/beta-1, alpha-4/beta-1, alpha-5/beta-1, alpha-8/beta-1, alpha-10/beta-1, alpha-11/beta-1 and alpha-V/beta-1 are receptors for fibronectin. Alpha-4/beta-1 recognizes one or more domains within the alternatively spliced CS-1 and CS-5 regions of fibronectin. Integrin alpha-5/beta-1 is a receptor for fibrinogen. Integrin alpha-1/beta-1, alpha-2/beta-1, alpha-6/beta-1 and alpha-7/beta-1 are receptors for laminin. Integrin alpha-4/beta-1 is a receptor for VCAM1. It recognizes the sequence Q-I-D-S in VCAM1. Integrin alpha-9/beta-1 is a receptor for VCAM1, cytotactin and osteopontin. It recognizes the sequence A-E-I-D-G-I-E-L in cytotactin. Integrin alpha-3/beta-1 is a receptor for epiligrin, thrombospondin and CSPG4. Alpha-3/beta-1 may mediate with LGALS3 the stimulation by CSPG4 of endothelial cells migration. Integrin alpha-V/beta-1 is a receptor for vitronectin. Beta-1 integrins recognize the sequence R-G-D in a wide array of ligands. Isoform 2 interferes with isoform 1 resulting in a dominant negative effect on cell adhesion and migration (in vitro). When associated with alpha-7/beta-1 integrin, regulates cell adhesion and laminin matrix deposition. Involved in promoting endothelial cell motility and angiogenesis. Involved in osteoblast compaction through the fibronectin fibrillogenesis cell-mediated matrix assembly process and the formation of mineralized bone nodules. May be involved in up-regulation of the activity of kinases such as PKC via binding to KRT1. Together with KRT1 and RACK1, serves as a platform for SRC activation or inactivation. Plays a mechanistic adhesive role during telophase, required for the successful completion of cytokinesis. Integrin alpha-3/beta-1 provides a docking site for FAP (seprase) at invadopodia plasma membranes in a collagen-dependent manner and hence may participate in the adhesion, formation of invadopodia and matrix degradation processes, promoting cell invasion. ITGA4:ITGB1 binds to fractalkine (CX3CL1) and may act as its coreceptor in CX3CR1-dependent fractalkine signaling (PubMed:23125415, PubMed:24789099). ITGA4:ITGB1 and ITGA5:ITGB1 bind to PLA2G2A via a site (site 2) which is distinct from the classical ligand-binding site (site 1) and this induces integrin conformational

Isoform 5: Isoform 5 displaces isoform 1 in striated muscles.


Tissue specificity

Isoform 1 is widely expressed, other isoforms are generally coexpressed with a more restricted distribution. Isoform 2 is expressed in skin, liver, skeletal muscle, cardiac muscle, placenta, umbilical vein endothelial cells, neuroblastoma cells, lymphoma cells, hepatoma cells and astrocytoma cells. Isoform 3 and isoform 4 are expressed in muscle, kidney, liver, placenta, cervical epithelium, umbilical vein endothelial cells, fibroblast cells, embryonal kidney cells, platelets and several blood cell lines. Isoform 4, rather than isoform 3, is selectively expressed in peripheral T-cells. Isoform 3 is expressed in non-proliferating and differentiated prostate gland epithelial cells and in platelets, on the surface of erythroleukemia cells and in various hematopoietic cell lines. Isoform 5 is expressed specifically in striated muscle (skeletal and cardiac muscle).

Sequence similarities

Belongs to the integrin beta chain family.

Contains 1 VWFA domain.

Post-translational modifications

The cysteine residues are involved in intrachain disulfide bonds.

Cellular localization

Cell membrane, sarcolemma. Cell junction. In cardiac muscle, isoform 5 is found in costameres and intercalated disks and Cell membrane. Cell projection, invadopodium membrane. Cell projection, ruffle membrane. Recycling endosome. Melanosomes. Cleavage furrow. Cell projection, lamellipodium. Cell projection, ruffle. Cell junction, focal adhesion. Cell surface. Isoform 2 does not localize to focal adhesions. Highly enriched in stage 1 melanosomes. Located on plasma membrane of neuroblastoma NMB7 cells. In a lung cancer cell line, in prometaphase and metaphase, localizes diffusely at the membrane and in a few intracellular vesicles. In early telophase, detected mainly on the matrix-facing side of the cells. By mid-telophase, concentrated to the ingressing cleavage furrow, mainly to the basal side of the furrow. In late telophase, concentrated to the extending protrusions formed at the opposite ends of the spreading daughter cells, in vesicles at the base of the lamellipodia formed by the separating daughter cells. Colocalizes with ITGB1BP1 and metastatic suppressor protein NME2 at the edge or peripheral ruffles and lamellipodia during the early stages of cell spreading on fibronectin or collagen. Translocates from peripheral focal adhesions sites to fibrillar adhesions in a ITGB1BP1-dependent manner. Enriched preferentially at invadopodia, cell membrane protrusions that correspond to sites of cell invasion, in a collagen-dependent manner. Localized at plasma and ruffle membranes in a collagen-independent manner.

Images
Lanes 1, 5 and 9: Wild-type HAP1 cell lysate (20 µg)
Lanes 2, 6 and 10: Integrin beta 1 knockout HAP1 cell lysate (20 µg)
Lanes 3, 7 and 11: U87-MG cell lysate (20 µg)
Lanes 4, 8 and 12: A431 cell lysate (20 µg)
Lanes 1, 2, 3 and 4: Green signal from target – ab52971 observed at 140 kDa
Lanes 5, 6, 7 and 8: Red signal from loading control – ab8245 observed at 37 kDa
Lanes 9, 10, 11 and 12: Merged (red and green) signal

ab52971 was shown to specifically react with Integrin beta 1 in wild-type HAP1 cells. No band was observed when Integrin beta 1 knockout samples were examined. Wild-type and Integrin beta 1 knockout samples were subjected to SDS-PAGE. ab52971 and ab8245 (loading control to GAPDH) were diluted 1/10,000 and 1/2000 respectively and incubated overnight at 4°C. 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 1/10,000 dilution for 1 hour at room temperature before imaging.

ab52971 at 1/500 staining Integrin beta 1 antibody in human transitional cell carcinoma of bladder by immunohistochemistry (FFPE).

Immunohistochemical analysis of paraffin-embedded human transitional cell carcinoma of bladder tissue labeling Integrin beta 1 with ab52971 at 1/500 dilution followed by goat anti-rabbit IgG H&L (HRP) (ab97051, 1/500). Counter stained with hematoxylin.
Lane 1: Wild-type HAP1 cell lysate (20 µg)
Lane 2: Integrin beta 1 knockout HAP1 cell lysate (20 µg)
Lane 3: U87-MG cell lysate (20 µg)
Lane 4: A431 cell lysate (20 µg)
Lanes 1 - 4: Merged signal (red and green). Green - ab52971 observed at 140 kDa. Red signal from loading control – ab8245 observed at 37 kDa.
This western blot image is a comparison between ab52971 and a competitor’s top cited rabbit polyclonal antibody.

All lanes: Anti-Integrin beta 1 antibody [EP1041Y] (ab52971) at 1/10000 dilution (purified)
Lane 1: U87-MG cell lysate
Lane 2: HT-1080 cell lysate
Lane 3: U937 cell lysate
Lysates/proteins at 20 µg per lane.

Secondary
All lanes: HRP goat anti-rabbit (H+L) at 1/1000 dilution

Predicted band size: 88 kDa
Observed band size: 140 kDa
why is the actual band size different from the predicted?

5% NFDM/TBST dilution buffer
All lanes: Anti-Integrin beta 1 antibody [EP1041Y] (ab52971) at 20 µg (unpurified)

Lane 1: HeLa (Human epithelial carcinoma cell line) Whole Cell Lysate
Lane 2: HT 1080 (Human fibrosarcoma) Whole Cell Lysate
Lane 3: U2OS (Human osteosarcoma cell line) Whole Cell Lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes: Goat Anti-Rabbit IgG H&L (Alexa Fluor® 790) (ab175781) at 1/10000 dilution

Predicted band size: 88 kDa
Observed band size: 120 + 140 kDa why is the actual band size different from the predicted?

This blot was produced using a 4-12% Bis-tris gel under the MOPS buffer system. The gel was run at 200V for 50 minutes before being transferred onto a Nitrocellulose membrane at 30V for 70 minutes. The membrane was then blocked for an hour using Licor blocking buffer before being incubated with ab52971 overnight at 4°C. Antibody binding was detected using ab175781 at a 1:10,000 dilution for 1hr at room temperature and then imaged using the Licor Odyssey CLx.

Secondary antibody - anti-rabbit Alexa Fluor 790
Unpurified ab52971 staining human breast cancer metastasis tissue sections by IHC-P. Sections were formaldehyde fixed and subjected to heat mediated antigen retrieval in citrate buffer (pH 6) prior to blocking with a commercial blocking reagent and incubation with the antibody (diluted 1/100) for 18 hours at 4°C. A HRP-conjugated goat anti-rabbit was used as the secondary antibody. This image shows a cancer metastasis at 40x with beta1 staining (in red) in both blood vessels and tumour cells. Blue is Hoechst for nuclei. The antibody detection was enhanced using a commercial Cy3 tyramide signal amplification kit.

Formaldehyde-fixed, paraffin-embedded human lung tissue stained for Integrin beta 1 using ab52971 at 1/100 dilution in immunohistochemical analysis.

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