abcam

Product datasheet

Anti-Integrin alpha 5 antibody [EPR7854] - Low endotoxin, Azide free ab221606





3 References 17 Images

Overview

Product name Anti-Integrin alpha 5 antibody [EPR7854] - Low endotoxin, Azide free

Rabbit monoclonal [EPR7854] to Integrin alpha 5 - Low endotoxin, Azide free **Description**

Host species Rabbit

Tested applications Suitable for: WB, IHC-P, ICC/IF, IP, Flow Cyt (Intra)

Species reactivity Reacts with: Mouse, Rat. Human

Immunogen Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

Positive control IHC-P: Human kidney, human cerebral cortex, mouse cerebral cortex, and rat cerebral cortex

tissues. ICC/IF: U937, MCF7 and wild-type HAP1 cells. Flow Cyt (intra): HeLa cells. IP: HeLa

cells.

General notes ab221606 is the carrier-free version of ab150361.

> Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency.

This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cellbased assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar[®] is a trademark of Fluidigm Canada Inc.

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

1

monoclonal antibodies. For details on our patents, please refer to **RabMAb**® **patents**.

Our <u>Low endotoxin, azide-free formats</u> have low endotoxin level (≤ 1 EU/ml, determined by the LAL assay) and are free from azide, to achieve consistent experimental results in functional assays.

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C. Do Not Freeze.

Storage buffer pH: 7.2

Constituent: PBS

Carrier free Yes

Purity Protein A purified

ClonalityMonoclonalClone numberEPR7854

Isotype IgG

Applications

The Abpromise guarantee Our <u>Abpromise guarantee</u> covers the use of ab221606 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		Use at an assay dependent concentration.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. See IHC antigen retrieval protocols
ICC/IF		Use at an assay dependent concentration.
IP		Use at an assay dependent concentration.
Flow Cyt (Intra)		Use at an assay dependent concentration. <u>ab199376</u> - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.

Target

Function Integrin alpha-5/beta-1 is a receptor for fibronectin and fibrinogen. It recognizes the sequence R-

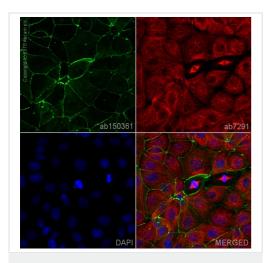
G-D in its ligands. In case of HIV-1 infection, the interaction with extracellular viral Tat protein

seems to enhance angiogenesis in Kaposi's sarcoma lesions.

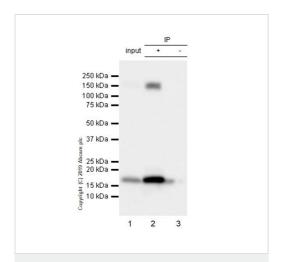
Sequence similarities Belongs to the integrin alpha chain family.

Contains 7 FG-GAP repeats.

Images



Immunocytochemistry/ Immunofluorescence - Anti-Integrin alpha 5 antibody [EPR7854] - Low endotoxin, Azide free (ab221606)



Immunoprecipitation - Anti-Integrin alpha 5 antibody [EPR7854] - Low endotoxin, Azide free (ab221606)

<u>ab150361</u> staining Integrin alpha 5 in MCF7 cells. The cells were fixed with 100% methanol (5min), permeabilized with 0.1% Tween for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with <u>ab150361</u> at 1μg/ml concentration and <u>ab7291</u> at 1μg/ml concentration overnight at +4°C, followed by a further incubation at room temperature for 1h with a goat secondary antibody to Rabbit lgG (Alexa Fluor® 488) (<u>ab150081</u>) at 2 μg/ml (shown in green) and a goat secondary antibody to Mouse lgG (Alexa Fluor® 594) (<u>ab150120</u>) at 2 μg/ml (showed in pseudocolour red). Nuclear DNA was labelled in blue with DAPI.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab150361</u>).

 $\underline{ab150361} \ (\text{Purified}) \ \text{at 1:20 dilution} \ (1 \ \mu g) \ \text{immunoprecipitating} \\ \text{Integrin alpha 5 in HeLa whole cell lysate}.$

Lane 1 (input): HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate 10 µg

Lane 2 (+): ab150361 & HeLa whole cell lysate

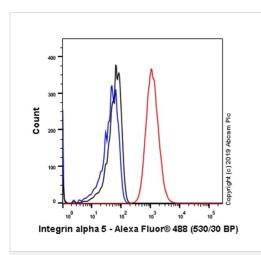
Lane 3 (-): Rabbit monoclonal IgG (ab172730) instead of

ab150361 in HeLa whole cell lysate

For western blotting, VeriBlot for IP Detection Reagent (HRP) (ab131366) was used at 1:5000 dilution.

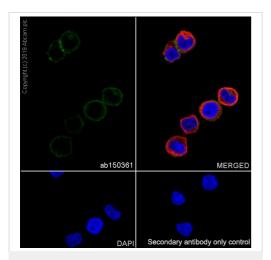
Blocking and diluting buffer: 5% NFDM/TBST.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab150361)



Flow Cytometry (Intracellular) - Anti-Integrin alpha 5 antibody [EPR7854] - Low endotoxin, Azide free (ab221606)

Intracellular Flow Cytometry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling Integrin alpha 5 with purified ab150361 at 1/20 dilution (10 µg/ml) (Red). Cells were fixed with 4% Paraformaldehyde and permeabilised with 90% Methanol. A Goat anti rabbit IgG (Alexa Fluor[®] 488, ab150077) secondary antibody was used at 1/2000. Isotype control - Rabbit monoclonal IgG (Black). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue). This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab150361)



Immunocytochemistry/ Immunofluorescence - Anti-Integrin alpha 5 antibody [EPR7854] - Low endotoxin, Azide free (ab221606)

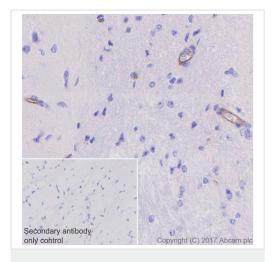
Immunocytochemistry/ Immunofluorescence analysis of U937 (Human histiocytic lymphoma monocyte) cells labeling Integrin alpha 5 with purified <u>ab150361</u> at 1:50 dilution (4.3 μg/ml). Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% tritonX-100. Cells were counterstained with Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1:200 (2.5 μg/ml). Goat anti rabbit lgG (Alexa Fluor® 488, <u>ab150077</u>) was used as the secondary antibody at 1:1000 (2 μg/ml) dilution. DAPI (blue) was used as nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control. This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab150361</u>)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Integrin alpha 5 antibody [EPR7854] - Low endotoxin, Azide free (ab221606)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of rat cerebral cortex tissue sections labeling Integrin alpha 5 with purified ab150361 at 1:400 dilution (0.54 µg/ml). Heat mediated antigen retrieval was performed using ab93684 (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.

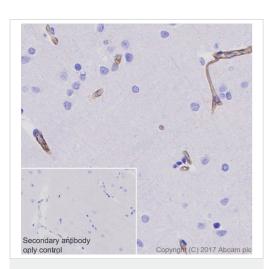
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab150361)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Integrin alpha 5 antibody [EPR7854] - Low endotoxin, Azide free (ab221606)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of mouse cerebral cortex tissue sections labeling Integrin alpha 5 with purified ab150361 at 1:400 dilution (0.54 µg/ml). Heat mediated antigen retrieval was performed using ab93684 (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.

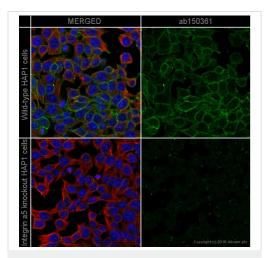
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab150361**)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Integrin alpha 5 antibody [EPR7854] - Low endotoxin, Azide free (ab221606)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human cerebral cortex tissue sections labeling Integrin alpha 5 with purified <u>ab150361</u> at 1:400 dilution (0.54 µg/ml). Heat mediated antigen retrieval was performed using <u>ab93684</u> (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.

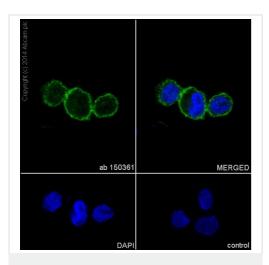
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab150361)



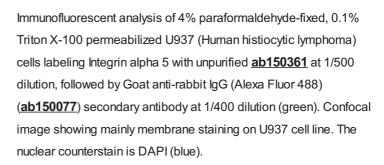
Immunocytochemistry/ Immunofluorescence - Anti-Integrin alpha 5 antibody [EPR7854] - Low endotoxin, Azide free (ab221606)

<u>ab150361</u> staining Integrin alpha 5 in wild-type HAP1 cells (top panel) and ITGA5 knockout HAP1 cells (bottom panel). The cells were fixed with 100% methanol (5min), permeabilized with 0.1% Tween for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with <u>ab150361</u> at 1/250 dilution and <u>ab7291</u> at 1μg/ml concentration overnight at +4°C, followed by a further incubation at room temperature for 1h with a goat secondary antibody to Rabbit lgG (Alexa Fluor® 488) (<u>ab150081</u>) at 2 μg/ml (shown in green) and a goat secondary antibody to Mouse lgG (Alexa Fluor® 594) (<u>ab150120</u>) at 2 μg/ml (showed in pseudocolour red) . Nuclear DNA was labelled in blue with DAPI.

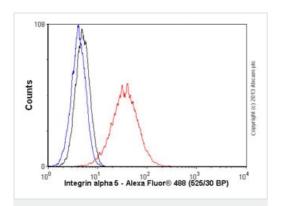
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab150361</u>).



Immunocytochemistry/ Immunofluorescence - Anti-Integrin alpha 5 antibody [EPR7854] - Low endotoxin, Azide free (ab221606)



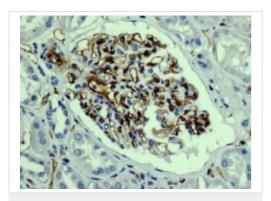
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab150361</u>).



Flow Cytometry (Intracellular) - Anti-Integrin alpha 5 antibody [EPR7854] - Low endotoxin, Azide free (ab221606)

Overlay histogram showing HeLa cells stained with unpurified ab150361 (red line). The cells were fixed with 4% paraformaldehyde (10 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 proteinprotein interactions followed by the antibody (ab150361, 1/1000 dilution) for 30 min at 22°C. The secondary antibody used was goat anti-rabbit Alexa Fluor 488 IgG (H&L) (ab150077) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit lgG (monoclonal) (0.1μg/1x10⁶ cells) used under the same conditions. Unlabelled sample (blue line) was also used as a control. Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter. This antibody gave a positive signal in HeLa cells fixed with 80% methanol (5 min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab150361).

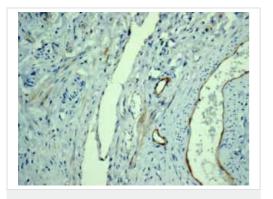


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Integrin alpha 5 antibody [EPR7854] - Low endotoxin, Azide free (ab221606)

Immunohistochemical analysis of paraffin embedded Human kidney tissue labelling Integrin alpha 5 with unpurified <u>ab150361</u> antibody at a dilution of 1/100.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab150361</u>).

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

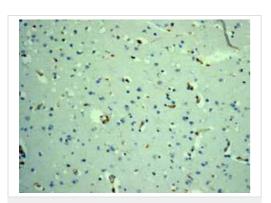


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Integrin alpha 5 antibody [EPR7854] - Low endotoxin, Azide free (ab221606)

Immunohistochemical analysis of paraffin embedded Human Vessels tissue using unpurified <u>ab150361</u> showing +ve staining.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab150361).

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

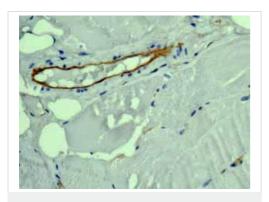


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Integrin alpha 5 antibody [EPR7854] - Low endotoxin, Azide free (ab221606)

Immunohistochemical analysis of paraffin embedded Human Brain vessels tissue using unpurified <u>ab150361</u> showing +ve staining.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab150361).

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

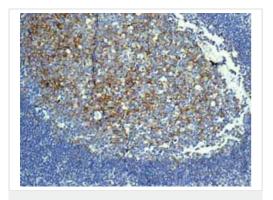


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Integrin alpha 5 antibody [EPR7854] - Low endotoxin, Azide free (ab221606)

Immunohistochemical analysis of paraffin embedded Human Skeletal muscle vessel tissue using unpurified <u>ab150361</u> showing +ve staining.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab150361</u>).

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

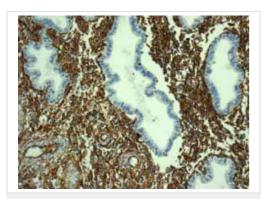


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Integrin alpha 5 antibody [EPR7854] - Low endotoxin, Azide free (ab221606)

Immunohistochemical analysis of paraffin embedded Normal Human Tonsil tissue using unpurified <u>ab150361</u> showing +ve staining.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab150361).

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

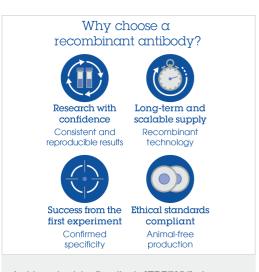


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Integrin alpha 5 antibody [EPR7854] - Low endotoxin, Azide free (ab221606)

Immunohistochemical analysis of paraffin embedded Normal Human Uterus tissue using unpurified <u>ab150361</u> showing +ve staining.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab150361</u>).

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Anti-Integrin alpha 5 antibody [EPR7854] - Low endotoxin, Azide free (ab221606)

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