abcam

Product datasheet

Anti-COL18A1 antibody [EPR23481-21] - BSA and Azide free ab275750



7 Images

Overview

Product name Anti-COL18A1 antibody [EPR23481-21] - BSA and Azide free

Description Rabbit monoclonal [EPR23481-21] to COL18A1 - BSA and Azide free

Host species Rabbit

Specificity This antibody does not recognise the Endostatin chain.

This antibody (ab275750 [EPR23481-21] is a distinct clone from ab275394 [EPR23481-240].

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

Species reactivity Reacts with: Human

Immunogen Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: HepG2 and Huh7 whole cell lysates. IHC-P: Human kidney and bladder cancer tissue.

ICC/IF: HepG2 cells. Flow Cyt (intra): HepG2 cells. IP: Huh7 whole cell lysate.

General notes ab275750 is the carrier-free version of ab275746.

> 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 monoclonal antibodies. For details on our patents, please refer to ${\hbox{\bf RabMAb}}^{\hbox{\bf @}}$ patents.

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C.

Storage buffer Constituent: 100% PBS

Carrier free Yes

Purity Protein A purified

Clonality Monoclonal
Clone number EPR23481-21

Isotype IgG

Applications

The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab275750 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 |
|------------------|-----------|---|
| IP | | Use at an assay dependent concentration. |
| ICC/IF | | Use at an assay dependent concentration. |
| IHC-P | | Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. |
| Flow Cyt (Intra) | | Use at an assay dependent concentration. |
| WB | | Use at an assay dependent concentration. Detects a band of approximately 178 kDa (predicted molecular weight: 178 kDa). |

Target

Function COLA18A probably plays a major role in determining the retinal structure as well as in the closure

of the neural tube.

Endostatin potently inhibits endothelial cell proliferation and angiogenesis. May inhibit

angiogenesis by binding to the heparan sulfate proteoglycans involved in growth factor signaling.

Tissue specificity Present in multiple organs with highest levels in liver, lung and kidney.

Involvement in disease Knobloch syndrome 1

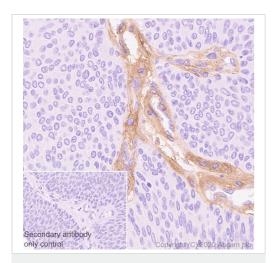
Sequence similarities Belongs to the multiplexin collagen family.

Contains 1 FZ (frizzled) domain.
Contains 1 Iaminin G-like domain.

Prolines at the third position of the tripeptide repeating unit (G-X-Y) of the triple-helical regions are hydroxylated.

Secreted, extracellular space, extracellular matrix.

Images



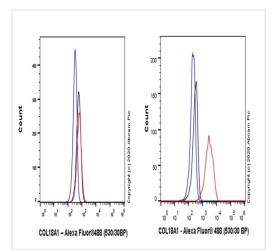
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-COL18A1 antibody
[EPR23481-21] - BSA and Azide free (ab275750)

This data was developed using <u>ab275746</u>, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded Human bladder cancer tissue labeling COL18A1 with <u>ab275746</u> at 1/4000 (0.134 ug/ml) dilution followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (<u>ab209101</u>). Positive staining on blood vessels of human bladder cancer (PMID:21815140). The section was incubated with <u>ab275746</u> for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND[®] RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins.



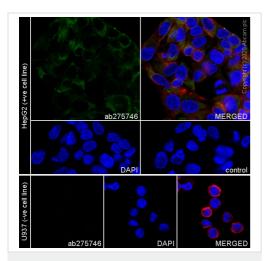
Flow Cytometry (Intracellular) - Anti-COL18A1 antibody [EPR23481-21] - BSA and Azide free (ab275750)

This data was developed using <u>ab275746</u>, the same antibody clone in a different buffer formulation.

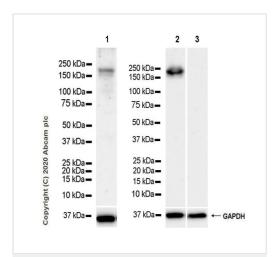
Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed, 90% methanol-permeabilized U-937 (Human histiocytic lymphoma monocyte, Left) / HepG2 (Human hepatocellular carcinoma epithelial cell, Right) cells labelling COL18A1 with ab275746 at 1/500 dilution (0.1ug) (Red) compared with a Rabbit monoclonal lgG (ab172730) isotype control (Black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue).

A Goat anti rabbit lgG (Alexa Fluor[®]488, <u>ab150077</u>) at 1/2000 dilution was used as the secondary antibody.

Negative control: U937 cell (PMID: 15451191).



Immunocytochemistry/ Immunofluorescence - Anti-COL18A1 antibody [EPR23481-21] - BSA and Azide free (ab275750)



Western blot - Anti-COL18A1 antibody [EPR23481-21] - BSA and Azide free (ab275750)

This data was developed using <u>ab275746</u>, the same antibody clone in a different buffer formulation.

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized HepG2 cells labelling COL18A1 with ab275746 at 1/50 (8.26 ug/ml) dilution, followed by ab150077 Goat Anti-Rabbit lgG H&L (Alexa Fluor[®] 488) antibody at 1/1000 dilution (Green). Confocal image showing cytoplasmic staining in HepG2 cells. Negative control: U937 cell (PMID: 15451191). ab195889 Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor[®] 594) was used to counterstain tubulin at 1/200 2.5 dilution (Red). The Nuclear counterstain was DAPI (Blue).

Secondary antibody only control: Secondary antibody is <u>ab150077</u> Goat Anti-Rabbit IgG H&L (Alexa Fluor[®] 488) at 1/1000 dilution.

All lanes : Anti-COL18A1 antibody [EPR23481-21] (**ab275746**) at 1/1000 dilution

Lane 1 : HepG2 (human hepatocellular carcinoma epithelial cell), whole cell lysate at 20 µg

Lane 2 : Huh7 (human hepatocellar carcinoma epithelial cell), whole cell lysate at 40 μg

Lane 3 : U937 (human histiocytic lymphoma monocyte), whole cell lysate at 40 μg

Secondary

All lanes : Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated (ab97051) at 1/100000 dilution

Predicted band size: 178 kDa **Observed band size:** 178 kDa

This data was developed using <u>ab275746</u>, the same antibody clone in a different buffer formulation.

Blocking and diluting buffer and concentration: 5% NFDM/TBST.

The expression profile observed is consistent with what has been described in the literature (PMID: 11581192, 15451191).

Negative control: U937 (PMID:15451191).

The lanes 1 of blot were developed using a higher sensitivity ECL substrate.

Exposure time: Lane 1: 3 minutes Lane 2-3: 70 seconds.

250 kDa —
150 kDa —
100 kDa —
75 kDa —
37 kDa —
37 kDa —
25 kDa —
20 kDa —
11 kDa —
110 kDa —
12 kDa —
13 kDa —
14 kDa —
14 kDa —
14 kDa —
15 kDa —
16 kDa —
17 kDa —
18 kDa —
19 kDa —
11 kDa —

Immunoprecipitation - Anti-COL18A1 antibody
[EPR23481-21] - BSA and Azide free (ab275750)

This data was developed using <u>ab275746</u>, the same antibody clone in a different buffer formulation.

COL18A1 was immunoprecipitated from 0.35 mg Huh7 (human hepatocellular carcinoma epithelial cell), whole cell lysate with **ab275746** at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using **ab275746** at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**) was used at 1/5000 dilution.

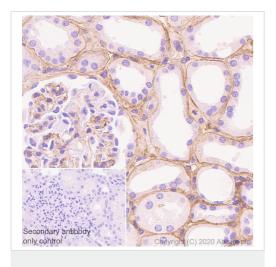
Lane 1: Huh7 (human hepatocellular carcinoma epithelial cell), whole cell lysate 10 ug

Lane 2: ab275746 IP in Huh7 whole cell lysate

Lane 3: Rabbit monoclonal $\lg G$ (<u>ab172730</u>) instead of 275746 in Huh7 whole cell lysate

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 3 minutes.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-COL18A1 antibody

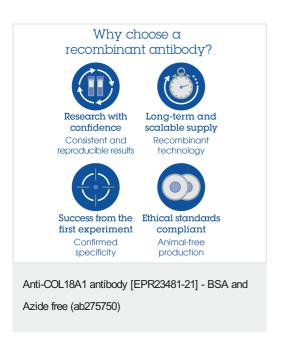
[EPR23481-21] - BSA and Azide free (ab275750)

This data was developed using <u>ab275746</u>, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded Human kidney tissue labeling COL18A1 with <u>ab275746</u> at 1/4000 (0.134 ug/ml) dilution followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (<u>ab209101</u>). Positive staining on human kidney (PMID: 12588956). The section was incubated with <u>ab275746</u> for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND[®] RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins.



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