abcam

Product datasheet

Anti-Fibronectin antibody [F1] - Low endotoxin, Azide free ab219366





Overview

Product name Anti-Fibronectin antibody [F1] - Low endotoxin, Azide free

Description Rabbit monoclonal [F1] to Fibronectin - Low endotoxin, Azide free

Host species Rabbit

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

Species reactivity Reacts with: Human

Immunogen Recombinant full length protein. This information is proprietary to Abcam and/or its suppliers.

Positive control Human serum and stomach tissue. This antibody gave a positive result in IF/ICC when used in the

following formaldehyde fixed cell lines: HepG2. Flow Cyt (intra): HepG2

General notes ab219366 is the carrier-free version of ab32419.

> 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 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 **RabMAb® patents**.

Our Low endotoxin, azide-free formats 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.

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

Properties

Form Liquid

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

Storage buffer pH: 7.20

Constituent: PBS

Carrier free Yes

Purity Protein A purified

Clonality Monoclonal

Clone number F1
Isotype IgG

Applications

The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab219366 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
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.
WB		Use at an assay dependent concentration. Predicted molecular weight: 263 kDa.
ICC/IF		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

Fibronectins bind cell surfaces and various compounds including collagen, fibrin, heparin, DNA, and actin. Fibronectins are involved in cell adhesion, cell motility, opsonization, wound healing, and maintenance of cell shape. Involved in osteoblast compaction through the fibronectin fibrillogenesis cell-mediated matrix assembly process, essential for osteoblast mineralization. Participates in the regulation of type I collagen deposition by osteoblasts.

Anastellin binds fibronectin and induces fibril formation. This fibronectin polymer, named superfibronectin, exhibits enhanced adhesive properties. Both anastellin and superfibronectin inhibit tumor growth, angiogenesis and metastasis. Anastellin activates p38 MAPK and inhibits lysophospholipid signaling.

Tissue specificity Plasma FN (soluble dimeric form) is secreted by hepatocytes. Cellular FN (dimeric or cross-

linked multimeric forms), made by fibroblasts, epithelial and other cell types, is deposited as fibrils

in the extracellular matrix. Ugl-Y1, Ugl-Y2 and Ugl-Y3 are found in urine.

Involvement in disease Glomerulopathy with fibronectin deposits 2

Sequence similaritiesContains 12 fibronectin type-I domains.

Contains 2 fibronectin type-II domains.

Contains 16 fibronectin type-III domains.

Developmental stage Ugl-Y1, Ugl-Y2 and Ugl-Y3 are present in the urine from 0 to 17 years of age.

Post-translational Sulfated.

modifications It is not known whether both or only one of Thr-2064 and Thr-2065 are/is glycosylated.

Forms covalent cross-links mediated by a transglutaminase, such as F13A or TGM2, between a

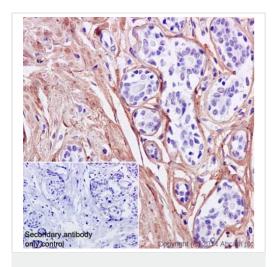
glutamine and the epsilon-amino group of a lysine residue, forming homopolymers and heteropolymers (e.g. fibrinogen-fibronectin, collagen-fibronectin heteropolymers).

Phosphorylated by FAM20C in the extracellular medium.

Proteolytic processing produces the C-terminal NC1 peptide, anastellin.

Cellular localization Secreted, extracellular space, extracellular matrix.

Images

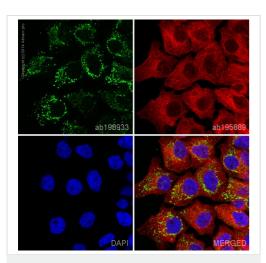


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Fibronectin antibody [F1]

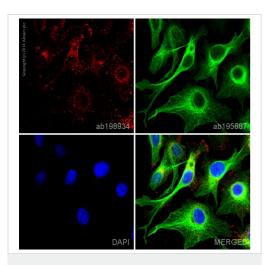
- Low endotoxin, Azide free (ab219366)

Immunohistochemical staining of paraffin embedded human breast carcinoma with purified <u>ab32419</u> at a dilution of 1/250. The secondary antibody used is <u>ab97051</u>, a HRP-conjugated goat antirabbit lgG (H+L), at a dilution of 1/500. The sample is counterstained with hematoxylin. Antigen retrieval was performed using Tris-EDTA buffer, pH 9.0. PBS was used instead of the primary antibody as the negative control, and is shown in the inset.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab32419).



Immunocytochemistry/ Immunofluorescence - Anti-Fibronectin antibody [F1] - Low endotoxin, Azide free (ab219366)



Immunocytochemistry/ Immunofluorescence - Anti-Fibronectin antibody [F1] - Low endotoxin, Azide free (ab219366)

Clone F1 (ab219366) has been successfully conjugated by Abcam. This image was generated using Anti-Fibronectin antibody [F1] (Alexa Fluor® 488). Please refer to **ab198933** for protocol details.

<u>ab198933</u> staining Fibronectin in A431 (Human epidermoid carcinoma cell line) cells. The cells were fixed with 4% formaldehyde (10 minutes), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1 hour. The cells were then incubated overnight at +4°C with <u>ab198933</u> at a 1/100 dilution (shown in green) and <u>ab195889</u>, Mouse monoclonal to alpha Tubulin (Alexa Fluor[®] 594), at a 1/250 dilution (shown in red).

Nuclear DNA was labelled with DAPI (shown in blue).

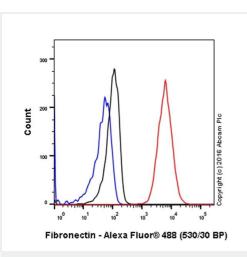
Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

Clone F1 (ab219366) has been successfully conjugated by Abcam. This image was generated using Anti-Fibronectin antibody [F1] (Alexa Fluor® 647). Please refer to **ab198934** for protocol details.

ab198934 staining Fibronectin in HepG2 cells. The cells were fixed with 4% formaldehyde (10 min), permeabilized with 0.1% Triton X-100 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 overnight at +4°C with **ab198934** at 1/100 dilution (shown in red) and **ab195887**, Mouse monoclonal to alpha Tubulin (Alexa Fluor[®] 488), at 1/250 dilution (shown in green). Nuclear DNA was labelled with DAPI (shown in blue).

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

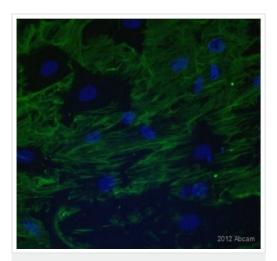
This product also gave a positive signal under the same testing conditions in HepG2 cells fixed with 100% methanol (5min).



Flow Cytometry (Intracellular) - Anti-Fibronectin antibody [F1] - Low endotoxin, Azide free (ab219366)

Intracellular Flow Cytometry analysis of HepG2 (Human liver hepatocellular carcinoma cell line) cells labeling Fibronectin with purified $\underline{ab32419}$ at 1/20 dilution (10 µg/mL) (red). Cells were fixed with 4% paraformaldehyde and permeabilized with 90% methanol. A Goat anti rabbit lgG (Alexa Fluor 488) at 1/2000 dilution was used as the secondary antibody. Rabbit monoclonal lgG (Black) was used as the isotype control, cells without incubation with primary antibody and secondary antibody (Blue) was used as the unlabeled control.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab32419).

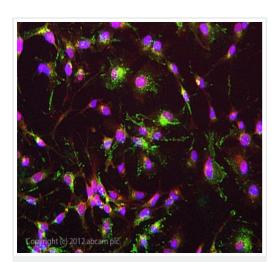


Immunocytochemistry/ Immunofluorescence - Anti-Fibronectin antibody [F1] - Low endotoxin, Azide free (ab219366)

This image is courtesy of an anonymous Abreview.

ICC/IF image of unpurified <u>ab32419</u> stained human mesenchymal stem cells. The cells were fixed in paraformaldehyde and then incubated in 0.1%BSA / 1% goat serum for 30 minutes, to block non-specific protein-protein interactions. The cells were then incubated with the antibody (<u>ab32419</u>, 1/100 dilution) for 2 hours at 22°C. The secondary antibody (green) was Alexa Fluor[®] 488 goat anti-rabbit IgG. DAPI was used to stain the cell nuclei (blue).

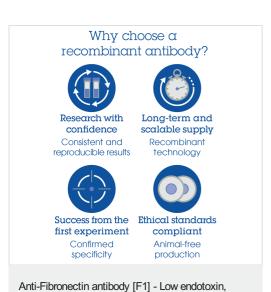
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab32419).



Immunocytochemistry/ Immunofluorescence - Anti-Fibronectin antibody [F1] - Low endotoxin, Azide free (ab219366)

ICC/IF image of unpurified <u>ab32419</u> stained HepG2 (Human liver hepatocellular carcinoma cell line) cells. The cells were fixed in 4% formaldehyde (10 minutes) and then incubated in 1%BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1 hour to permeabilize the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody <u>ab32419</u> at 1/100 dilution overnight at +4°C. The secondary antibody (green) was DyLight[®] 488 goat anti- rabbit (<u>ab96899</u>) lgG (H+L) used at a 1/1000 dilution for 1 hour. Alexa Fluor[®] 594 WGA was used to label plasma membranes (red) at a 1/200 dilution for 1 hour. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43 μ M.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab32419).



Azide free (ab219366)

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