abcam

Product datasheet

Anti-Fibronectin antibody [EPR23110-46] ab268020

KO VALIDATED Recombinant RabMAb

★★★★★ 1 Abreviews 22 References 12 Images

Overview

Product name	Anti-Fibronectin antibody [EPR23110-46]
Description	Rabbit monoclonal [EPR23110-46] to Fibronectin
Host species	Rabbit
Tested applications	**Suitable for:** IHC-P, IHC-Fr, ICC/IF, WB **Unsuitable for:** Flow Cyt or IP
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: Human serum, kidney and stomach lysate. Mouse, human and rat plasma lysate. NIH/3T3 whole cell lysate; HAP1 whole cell lysate. IHC-P: Human mammary gland tissue. Human esophageal squamous cell carcinoma tissue. Mouse and rat stomach tissue human pancreatic carcinoma tissue. ICC/IF: HepG2 and NIH/3T3 cells.
General notes	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**.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle.
Storage buffer	pH: 7.2 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal

Clone number	EPR23110-46
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab268020 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		1/2000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
IHC-Fr	★★★★★ (1)	1/500. Heat mediated antigen retrieval using sodium citrate buffer (10mM citrate pH 6.0 + 0.05% Tween-20).
ICC/IF		1/50.
WB		1/1000. Detects a band of approximately 285 kDa (predicted molecular weight: 262 kDa). This antibody can be used at 0.585 µg/ml.

Application notes Is unsuitable for Flow Cyt or IP.

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

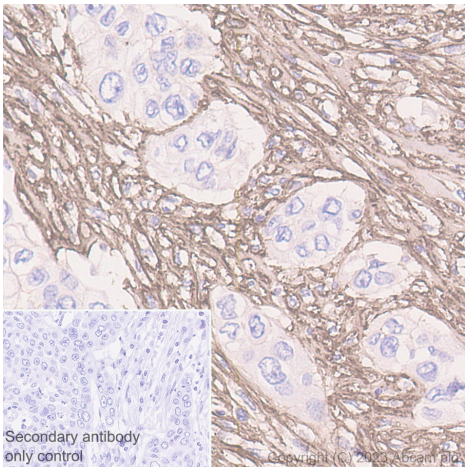
Sequence similarities Contains 12 fibronectin type-I domains.
Contains 2 fibronectin type-II domains.
Contains 16 fibronectin type-III domains.

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

Post-translational modifications Sulfated.
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

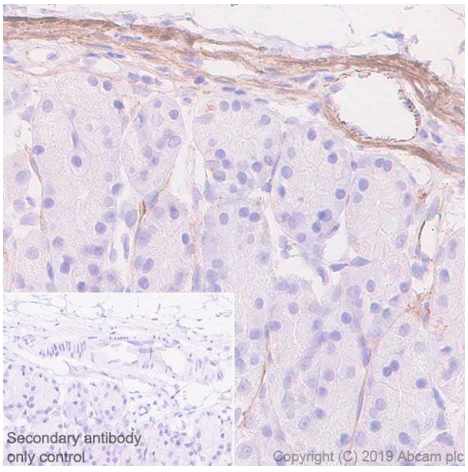


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Fibronectin antibody [EPR23110-46] (ab268020)

Immunohistochemical analysis of paraffin-embedded Human pancreatic carcinoma tissue labeling Fibronectin with ab268020 at 1/4000 dilution (0.146 µg/ml) followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP). The section was incubated with ab268020 at 4°C overnight. Positive staining in the human pancreatic carcinoma counterstained with hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP)(**ab214880**)

Heat mediated antigen retrieval using **ab93684** (Tris/EDTA buffer, pH 9.0).

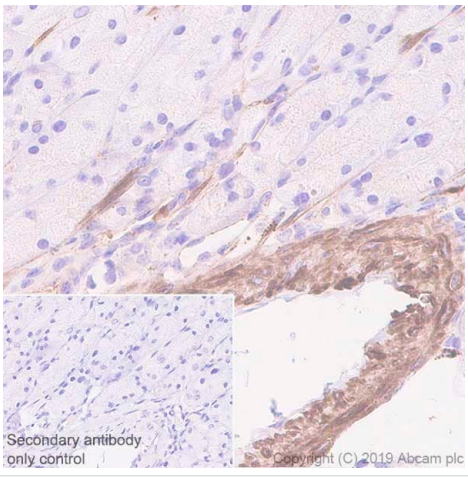


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Fibronectin antibody [EPR23110-46] (ab268020)

Immunohistochemical analysis of paraffin-embedded rat stomach tissue labeling Fibronectin with ab268020 at 1/2000 dilution (0.29 µg/ml) followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP). Positive staining in the smooth muscle of rat stomach (PMID:650101) Counterstained with hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP).

Heat mediated antigen retrieval using **ab93684** (Tris/EDTA buffer, pH 9.0).

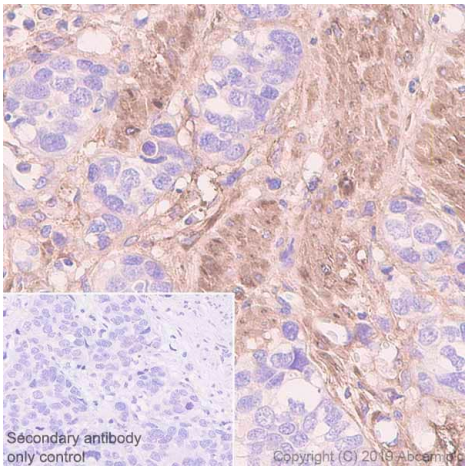


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Fibronectin antibody [EPR23110-46] (ab268020)

Immunohistochemical analysis of paraffin-embedded mouse stomach tissue labeling Fibronectin with ab268020 at 1/2000 dilution (0.29 µg/ml) followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP). Positive staining in the smooth muscle of mouse stomach (PMID:650101) Counterstained with hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP).

Heat mediated antigen retrieval using **ab93684** (Tris/EDTA buffer, pH 9.0).

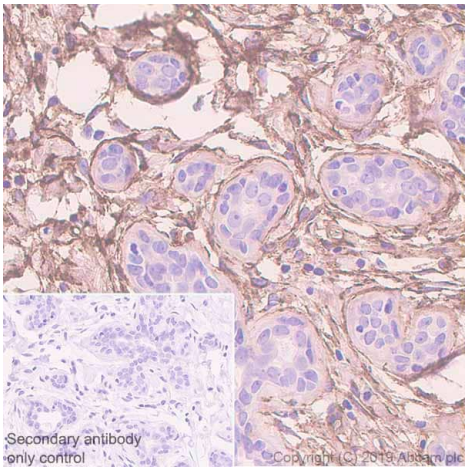


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Fibronectin antibody [EPR23110-46] (ab268020)

Immunohistochemical analysis of paraffin-embedded human esophageal squamous cell carcinoma tissue labeling Fibronectin with ab268020 at 1/2000 dilution (0.29 µg/ml) followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP). Positive staining in the stroma of human esophageal squamous cell carcinoma (PMID:30314454). Counterstained with hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP).

Heat mediated antigen retrieval using **ab93684** (Tris/EDTA buffer, pH 9.0).

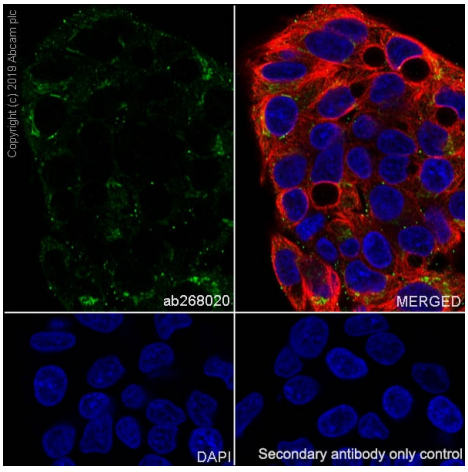


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Fibronectin antibody [EPR23110-46] (ab268020)

Immunohistochemical analysis of paraffin-embedded human mammary gland tissue labeling Fibronectin with ab268020 at 1/2000 dilution (0.29 µg/ml) followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP). Positive staining in the stroma of human mammary gland (PMID:650101). Counterstained with hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP).

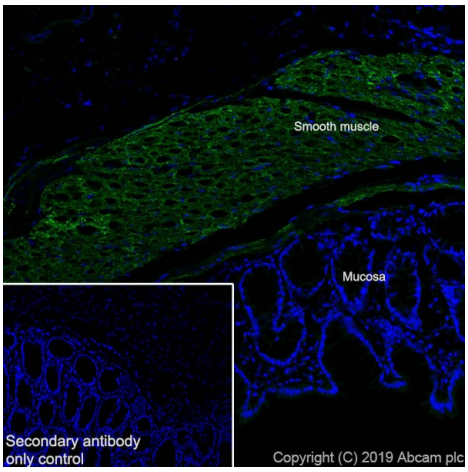
Heat mediated antigen retrieval using **ab93684** (Tris/EDTA buffer, pH 9.0).



Immunocytochemistry/ Immunofluorescence - Anti-Fibronectin antibody [EPR23110-46] (ab268020)

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized HepG2 (human hepatocellular carcinoma epithelial cell) cells labeling Fibronectin with ab268020 at 1/50 dilution, followed by **ab150077** AlexaFluor®488 Goat anti-Rabbit secondary antibody at 1/1000 dilution (Green). Confocal image showing cytoplasmic staining in HepG2 cells is observed. **ab195889** Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) was used to counterstain tubulin at 1/200 dilution (Red). The nuclear counterstain was DAPI (Blue).

Secondary antibody only control: Secondary antibody is **ab150077** AlexaFluor®488 Goat anti-Rabbit secondary at 1/1000 dilution.

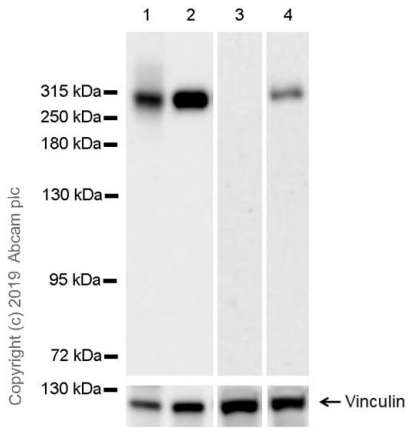


Immunohistochemistry (Frozen sections) - Anti-Fibronectin antibody [EPR23110-46] (ab268020)

Immunohistochemical analysis of frozen section of 4% PFA-fixed, 0.2% Triton X-100 permeabilized rat colon tissue labeling Fibronectin with ab268020 at 1/100 dilution followed by **ab150077** AlexaFluor®488 Goat anti-Rabbit secondary at 1/1000 dilution (Green). Positive staining mainly on smooth muscle of rat colon (PMID: 17881565) is observed. The nuclear counterstain is DAPI (Blue).

Secondary antibody only control: Secondary antibody is **ab150077** AlexaFluor®488 Goat anti-Rabbit secondary at 1/1000 dilution

Heat mediated antigen retrieval using sodium citrate buffer (10mM citrate pH 6.0 + 0.05% Tween-20).



Western blot - Anti-Fibronectin antibody [EPR23110-46] (ab268020)

All lanes : Anti-Fibronectin antibody [EPR23110-46] (ab268020) at 0.585 µg/ml (1:1000 dilution)

Lane 1 : Human stomach tissue lysate at 10 µg
Lane 2 : NIH/3T3 (mouse embryonic fibroblast), whole cell lysate at 10 µg
Lane 3 : Fibronectin-1 knockout HAP1 whole cell lysate at 40 µg
Lane 4 : Wild-type HAP1 whole cell lysate at 40 µg

Developed using the ECL technique.

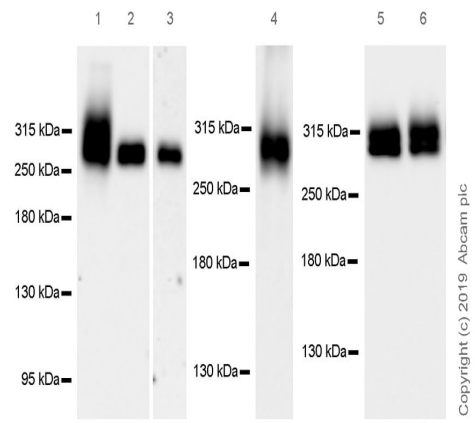
Predicted band size: 262 kDa
Observed band size: 285 kDa

ab268020 was shown to specifically react with Fibronectin in wild-type HAP1 cells as signal was lost in Fibronectin knockout cells. Wild-type and Fibronectin knockout samples were subjected to SDS-PAGE. ab268020 and **ab129002** (Rabbit anti-Vinculin loading control) were incubated 1 hour at room temperature at 1/1000 dilution and 1/5000 dilution respectively. Blots were developed with Goat Anti-Rabbit IgG, (H+L), Peroxidase

conjugated (**ab97051**) secondary antibody at 1/100,000 dilution for 1 hour at room temperature before imaging. The blot was developed on a BIO-RAD® ChemiDoc™ MP instrument using the ECL technique.

Exposure time: Lanes 1-2: 37 seconds; Lanes 3-4: 3 minutes.

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



Western blot - Anti-Fibronectin antibody [EPR23110-46] (ab268020)

All lanes : Anti-Fibronectin antibody [EPR23110-46] (ab268020) at 1/1000 dilution

Lane 1 : Human serum
Lane 2 : Human plasma
Lane 3 : Human kidney tissue lysate
Lane 4 : Human stomach tissue lysate
Lane 5 : Mouse plasma
Lane 6 : Rat plasma

Lysates/proteins at 20 µg per lane.

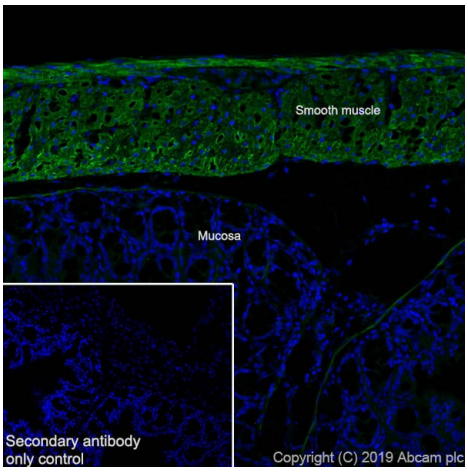
Secondary
Lanes 1-4 : VeriBlot for IP Detection Reagent (HRP) (**ab131366**) at 1/1000 dilution
Lanes 5-6 : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/100000 dilution

Predicted band size: 262 kDa
Observed band size: 285 kDa

Blocking and dilution buffer: 5% NFDM/TBST.

Exposure times: Lanes 1-2, 5-6: 3 seconds; Lane 3: 125 seconds; Lane 4: 33 seconds.

The expression molecular weight observed is consistent with what has been described in the literature (PMID:28367057).



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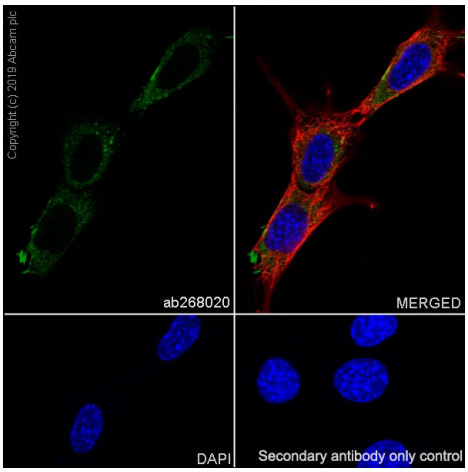

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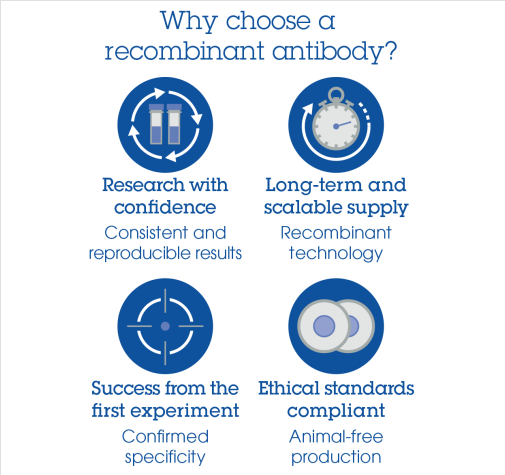

Anti-Fibronectin antibody [EPR23110-46] (ab268020)

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