

Fibronectin Polyclonal Antibody

Catalog NumberPA1-23693

Product data sheet

Details		Species Reactivity	
Size	500 µL	Species reactivity	Human, Pig
Host/Isotope	Rabbit / IgG	Published species	Pig, Sheep, Mouse, Not Applicable
Class	Polyclonal	Tested Applications	
Type	Antibody	Immunohistochemistry (Paraffin) (IHC (P))	1:100
Immunogen	Highly purified, human fibronectin from plasma.	Western Blot (WB)	2 µg/mL
Conjugate	Unconjugated	Immunocytochemistry (ICC/IF)	1:100
Form	Liquid	Published Applications	
Concentration	2 mg/mL	ELISA (ELISA)	See 1 publications below
Purification	Protein A	Immunocytochemistry (ICC/IF)	See 1 publications below
Storage buffer	PBS, pH 7.4, with 1% BSA	Immunohistochemistry (IHC)	See 2 publications below
Contains	0.05% sodium azide	* Suggested working dilutions are given as a guide only. It is recommended that the user titrate the product for use in their own experiment using appropriate negative and positive controls.	
Storage Conditions	4° C, do not freeze		

Product specific information

This antibody does not cross-react with Fibrinogen or other serum proteins. Prolonged fixation in buffered formalin may destroy the epitope. Auto/Zyme predigestion is recommended when using this antibody with formalin-fixed paraffin-embedded tissue sections. Suggested positive controls for this product are human tonsil, liver, skin or kidney.

Background/Target Information

Fibronectin is a disulfide-bonded dimer with a molecular weight range of 230-250 kDa. In the extracellular matrix of several connective tissues and vessels, fibronectin is present as an insoluble protein that is extensively cross-linked by interchain disulfide bonds forming high molecular mass polymers. Fibronectin is most abundant during embryonic development and tissue remodeling. Fibronectin is also present at high concentrations as a soluble plasma protein. Fibronectin is organized as a linear series of repeating modules which form domains for interaction with fibronectin itself, other matrix components (e.g. collagen and heparin) and receptors on cells (e.g. integrins). Fibronectin is present in a soluble dimeric form in plasma, and in a dimeric or multimeric form at the cell surface and in extracellular matrix. Fibronectin is involved in cell adhesion and migration processes including embryogenesis, wound healing, blood coagulation, host defense, and metastasis. Fibronectin has been implicated in carcinoma development in lung cancer. Further, Fibronectin expression is increased especially in non-small cell lung carcinoma. The adhesion of lung carcinoma cells to Fibronectin enhances tumorigenicity and confers resistance to apoptosis induced by standard chemotherapeutic agents. The gene encoding Fibronectin has three regions subject to alternative splicing, with the potential to produce 20 different transcript variants. However, the full-length nature of some Fibronectin variants has not been determined.

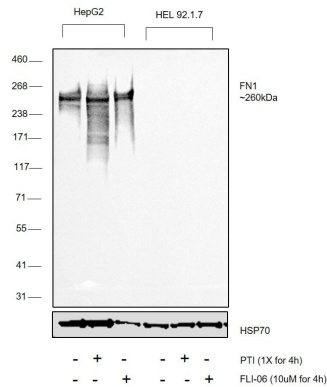
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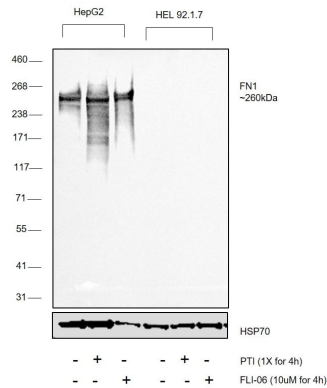
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Product Images For Fibronectin Polyclonal Antibody



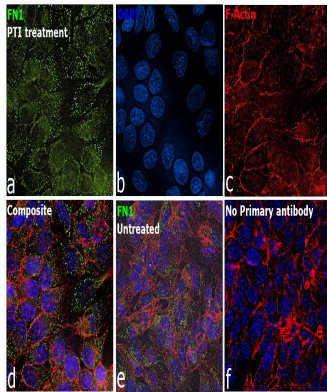
Fibronectin Antibody (PA1-23693) in WB

Western blot was performed using Anti-Fibronectin Polyclonal Antibody (Product # PA1-23693) and a ~260 kDa band corresponding to Fibronectin was observed across cell lines tested. Whole cell extracts (30 µg lysate) of Hep G2 (Lane 1), Hep G2 treated with 1X PTI for 4h (Lane 2), Hep G2 treated with 10 µm FLI-06 for 4h (Lane 3), HEL 92.1.7 (Lane 4), HEL 92.1.7 treated with 1X PTI for 4h (Lane 5), HEL 92.1.7 treated with 10 µm FLI-06 for 4h (Lane 6) were electrophoresed using NuPAGE™ 3-8% Tris-Acetate Protein Gel (Product # EA0378BOX). The blot was probed with the primary antibody (2 µg/mL) and detected by chemiluminescence with Goat anti-Rabbit IgG (Heavy Chain) Superclonal™ Recombinant Secondary Antibody, HRP (Product # A27036, 1:4000) using the iBright FL 1000 (Product # A32752). Chemiluminescent detection was performed using Novex® ECL Chemiluminescent Substrate Reagent Kit (Product # WP20005). Anti-Fibronectin Polyclonal Antibody (Product # PA1-23693) showed enhanced pick up in positive cell line Hep G2 upon treatment with secretion blockers as compared to in negative cell line HEL 92.1.7 treated with the same blockers.



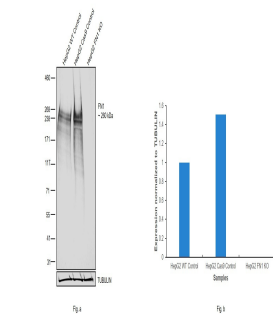
Fibronectin Antibody (PA1-23693)

Antibody specificity was demonstrated by detection of differential basal expression of the target across cell lines owing to their inherent genetic constitution. Relative expression of Fibronectin was observed in HepG2 upon protein transport inhibitors 1X PTI for 4h and 10uM FLI-06 for 4h versus in HEL 9.2.17 using the same treatments, using Fibronectin Polyclonal Antibody (Product # PA1-23693) in Western Blot. {TM}



Fibronectin Antibody (PA1-23693) in ICC/IF

Immunofluorescence analysis of Fibronectin was performed using 70% confluent log phase Hep G2 cells. The cells were fixed with 4% paraformaldehyde for 10 minutes, permeabilized with 0.1% Triton™ X-100 for 15 minutes, and blocked with 2% BSA for 45 minutes at room temperature. The cells were labeled with Fibronectin Monoclonal Antibody (3F12) (Product # MA5-14737) at 1:100 dilution in 0.1% BSA, incubated at 4 degree celsius overnight and then labeled with Donkey anti-Mouse IgG (H+L) Highly Cross-Adsorbed Secondary Antibody, Alexa Fluor Plus 488 (Product # A32766), (1:2000), for 45 minutes at room temperature (Panel a: Green). Nuclei (Panel b:Blue) were stained with ProLong™ Diamond Antifade Mountant with DAPI (Product # P36962). F-actin (Panel c: Red) was stained with Rhodamine Phalloidin (Product # R415, 1:300). Panel d represents the merged image showing cytosolic localization. Panel e represents untreated Hep G2 cells with no expression. Panel f represents control cells with no primary antibody to assess background The images were captured at 60X magnification. Cells treated with 1X PTI for 4h (Panel a) showed enhanced expression as compared to untreated cells (Panel e).



Fibronectin Antibody (PA1-23693)

Antibody specificity was demonstrated by CRISPR-Cas9 mediated knockout of target protein. A loss of signal was observed for target protein inFN1 KO cell line compared to control cell line using a Fibronectin Polyclonal Antibody (Product # PA1-23693). {KO}

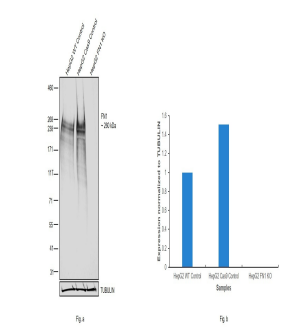
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Fibronectin Antibody (PA1-23693) in WB

Knockout of FN1 was achieved by CRISPR-Cas9 genome editing using LentiArray™ Lentiviral sgRNA (Product # A32042, Assay ID CRISPR616644_LV) and LentiArray Cas9 Lentivirus (Product # A32064). Western blot analysis of FN1 was performed by loading 30 µg of HepG2 Wild Type (Lane 1), HepG2 Cas9 (Lane 2) and HepG2 FN1 KO (Lane 3) whole cell extracts. The samples were electrophoresed using NuPAGE™ Novex™ 4-12% Bis-Tris Protein Gel (Product # NP0322BOX). Resolved proteins were then transferred onto a nitrocellulose membrane (Product # IB23001) by iBlot® 2 Dry Blotting System (Product # IB21001). The blot was probed with Fibronectin Polyclonal Antibody (Product # PA1-23693, 2 µg/mL dilution) and Goat anti-Rabbit IgG (Heavy Chain) Superclonal™ Recombinant Secondary Antibody, HRP (Product # A27036, 1:6,000 dilution) using the iBright FL 1000 (Product # A32752). Chemiluminescent detection was performed using Novex® ECL Chemiluminescent Substrate Reagent Kit (Product # WP20005). Loss of signal upon CRISPR mediated knockout (KO) using the LentiArray™ CRISPR product line confirms that antibody is specific to FN1.



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PubMed References For Fibronectin Polyclonal Antibody	
1 ELISA References	
Species / Dilution	Summary
Not Applicable / 1:1000	PA1-23693 was used in ELISA to learn about dependence on composition and assembly of endogenous extracellular matrices by blood-brain barrier properties in vitro
	Cell and tissue research (2016; 365: 233) "Blood-brain barrier properties in vitro depend on composition and assembly of endogenous extracellular matrices." Author(s):Zobel K,Hansen U,Galla HJ PubMed Article URL: http://dx.doi.org/10.1007/s00441-016-2397-7
1 Immunocytochemistry References	
Species / Dilution	Summary
Sheep / 1:400	PA123693 was used in immunocytochemistry to discuss the aggregative behavior and hair-inducing activity of ovine and human dermal papilla cells
	International journal of trichology (2016; 8: 121) "Characterization of Ovine Dermal Papilla Cell Aggregation." Author(s):Sari AR,Rufaut NW,Jones LN,Sinclair RD PubMed Article URL: http://dx.doi.org/10.4103/0974-7753.188966
2 Immunohistochemistry References	
Species / Dilution	Summary
Mouse / Not Cited	PA1-23693 was used in immunohistochemistry to investigate the strain difference between capsular and acapsular <i>Staphylococcus aureus</i> in the pathogenesis of brain abscess
	Journal of neuroimmunology (2010; 218: 83) "Evaluation of capsular and acapsular strains of S. aureus in an experimental brain abscess model." Author(s):Esen N,Wagoner G,Philips N PubMed Article URL: http://dx.doi.org/10.1016/j.jneuroim.2009.10.006
Pig / 1:100	PA1-23693 was used in immunohistochemistry to investigate the effect of simvastatin on renal fibrosis
	Journal of hypertension (2008; 26: 1651) "Simvastatin abates development of renal fibrosis in experimental renovascular disease." Author(s):Chade AR,Zhu XY,Grande JP,Krier JD,Lerman A,Lerman LO PubMed Article URL: http://dx.doi.org/10.1097/HJH.0b013e328302833a

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