


Product datasheet

Anti-Villin antibody [SP145] ab130751

Recombinant **RabMAb**

★★★★★ **4 Abreviews** **39 References** [7 Images](#)

Overview

Product name	Anti-Villin antibody [SP145]
Description	Rabbit monoclonal [SP145] to Villin
Host species	Rabbit
Tested applications	Suitable for: IP, mlHC, WB, IHC-P, Flow Cyt (Intra)
Species reactivity	Reacts with: Human Predicted to work with: Mouse, Chicken, Cow, Pig 
Immunogen	Synthetic peptide within Human Villin aa 800 to the C-terminus (C terminal). The exact sequence is proprietary. Database link: P09327
Positive control	Human colon, small intestine and kidney tissues; HT-29 cell lysate. mlHC: Human colon tissue. Flow cyto(intra): HT-29 cells.
General notes	This product is a recombinant monoclonal antibody, which offers several advantages including: <ul style="list-style-type: none"> - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production For more information see here . This product is FOR RESEARCH USE ONLY. For commercial use, please contact partnerships@abcam.com.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C. Do Not Freeze.
Storage buffer	pH: 7.60 Preservative: 0.1% Sodium azide Constituents: PBS, 1% BSA
Purity	Protein A/G purified
Purification notes	Purified from TCS by protein A/G.

Clonality	Monoclonal
Clone number	SP145
Isotype	IgG

Applications

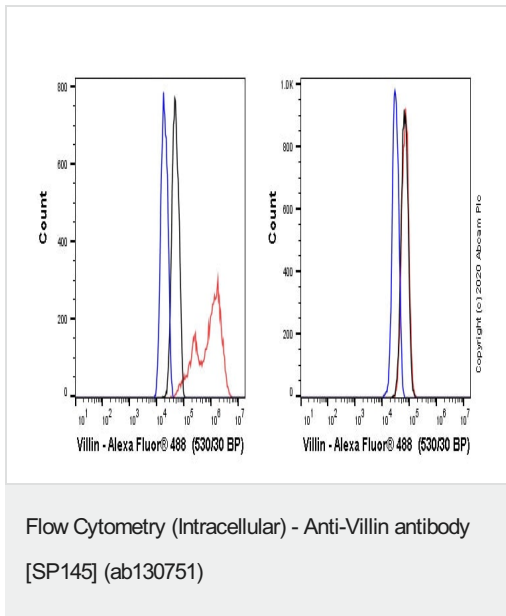
The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab130751 in the following tested applications. The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
IP		Use a concentration of 5 µg/ml.
mIHC		Use at an assay dependent concentration.
WB		1/100. Predicted molecular weight: 93 kDa. Incubate for 1 hour at room temperature.
IHC-P	★★★★★ (3)	1/100. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol. Primary incubation for 10 minutes at room temperature.
Flow Cyt (Intra)		1/100. ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

Target

Function	Ca(2+)-regulated actin-binding protein.
Tissue specificity	Major component of microvilli of intestinal epithelial cells and kidney proximal tubule cells.
Sequence similarities	Belongs to the villin/gelsolin family. Contains 6 gelsolin-like repeats. Contains 1 HP (headpiece) domain.
Domain	Consists of a large core fragment, the N-terminal portion, and a small headpiece, the C-terminal portion. The headpiece binds F-actin strongly in both the presence and absence of calcium.
Cellular localization	Cytoplasm > cytoskeleton.

Images



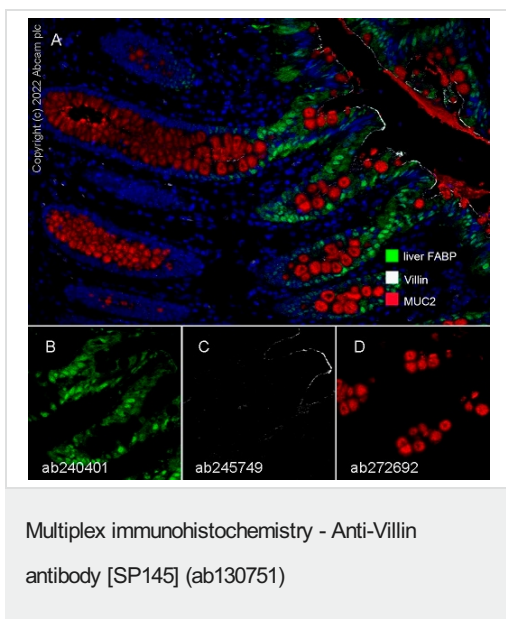
Flow cytometry overlay histogram showing left HT-29 positive cells and right negative PANC-1 stained with ab130751 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilised with 0.1% PBS-Triton X-100 for 15 min. The cells were then incubated in 1x PBS containing 10% normal goat serum to block non-specific protein-protein interaction followed by the antibody (ab130751) (1×10^6 in 100 μ l at 0.2 μ g/ml (1/1200)) for 30min at 22°C.

The secondary antibody Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed was incubated at 1/4000 for 30min at 22°C

Isotype control antibody (black line) was Recombinant Rabbit IgG, monoclonal [EPR25A] - Isotype Control used at the same concentration and conditions as the primary antibody. Unlabelled sample (blue line) was also used as a control.

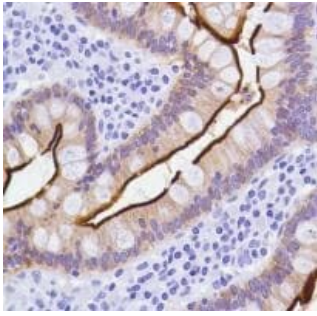
Acquisition of >5000 events were collected using a 50 mW Blue laser (488nm) and 525/40 bandpass filter.

This antibody gave a positive signal in HT-29 Fixed with 4% formaldehyde (10 min) / permeabilised with 0.1% PBS-Triton X-100 for 15 min under the same conditions.



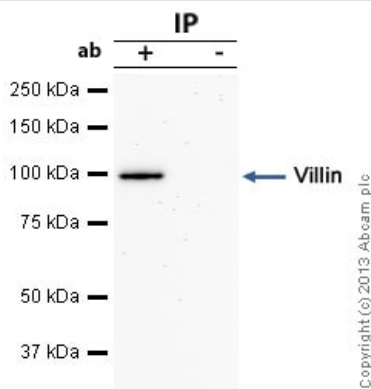
Fluorescence multiplex immunohistochemical analysis of the human colon (Formalin/PFA-fixed paraffin-embedded sections). Panel A: merged staining of anti-Villin (**ab245749**, gray; Opal™690), anti-liver FABP (**ab240401**, green; Opal™520) and anti-MUC2 (**ab272692**, red; Opal™570) on human colon. Panel B: anti-liver FABP stained on enterocytes. Panel C: anti-Villin stained on apical border. Panel D: anti-MUC2 stained on goblet cells. Opal Polymer HRP Ms + Rb was used as a secondary antibody. The immunostaining was performed on a Leica Biosystems BOND® RX instrument with an Opal™ 4-color kit. The section was incubated in three rounds of staining: in the order of **ab245749** (1/1000 dilution), **ab240401** (1/8000 dilution), and **ab272692** (1/5000 dilution) for 30 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins. DAPI (blue) was used as a nuclear counter

stain. Image acquisition was performed with Leica SP8 confocal microscope.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Villin antibody [SP145] (ab130751)

ab130751, at 1/100 dilution, staining Villin in formalin-fixed, paraffin-embedded Human colon tissue by Immunohistochemistry.



Immunoprecipitation - Anti-Villin antibody [SP145] (ab130751)

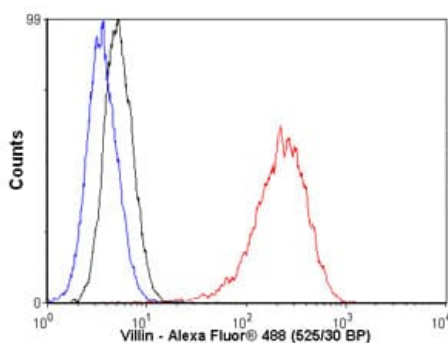
Villin was immunoprecipitated using 0.5mg SW480 whole cell extract, 5µg of Rabbit polyclonal to Villin and 50µl of protein G magnetic beads (+). No antibody was added to the control (-).

The antibody was incubated under agitation with Protein G beads for 10min, SW480 whole cell extract lysate diluted in RIPA buffer was added to each sample and incubated for a further 10min under agitation.

Proteins were eluted by addition of 40µl SDS loading buffer and incubated for 10min at 70°C; 10µl of each sample was separated on a SDS PAGE gel, transferred to a nitrocellulose membrane, blocked with 5% BSA and probed with ab130751.

Secondary: Mouse monoclonal [SB62a] Secondary Antibody to Rabbit IgG light chain (HRP) ([ab99697](#)).

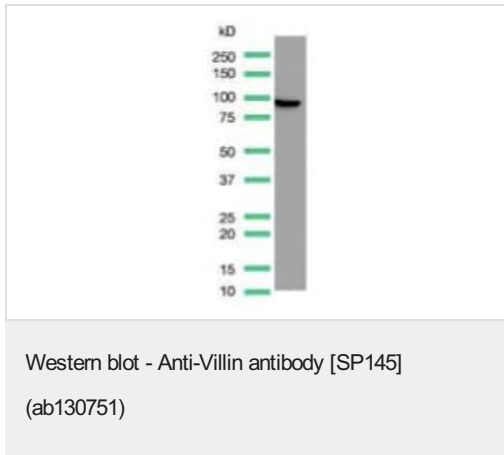
Band: 93kDa; Villin



Flow Cytometry (Intracellular) - Anti-Villin antibody [SP145] (ab130751)

Overlay histogram showing Caco 2 cells stained with ab130751 (red line). The cells were fixed with 80% methanol (5 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 protein-protein interactions followed by the antibody (ab130751, 1/100 dilution) for 30 min at 22°C. The secondary antibody used was Alexa Fluor® 488 goat anti-rabbit IgG (H&L) ([ab150077](#)) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) (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.



Anti-Villin antibody [SP145] (ab130751) at 1/100 dilution + HT-29 cell lysate

Predicted band size: 93 kDa

Why choose a recombinant antibody?

 <p>Research with confidence Consistent and reproducible results</p>	 <p>Long-term and scalable supply Recombinant technology</p>
 <p>Success from the first experiment Confirmed specificity</p>	 <p>Ethical standards compliant Animal-free production</p>

Anti-Villin antibody [SP145] (ab130751)

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