abcam

ab52640

Anti-VAV2 antibody [EP1067Y]

Recombinant | RabMAb | KO Validated | 20ul selling size

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Preservative: 0.05% Sodium azide Constituents: 50% Tissue culture supernatant, 40% Glycerol (glycerin, glycerine), 9.85% Tris glycine, 0.1% BSA
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Clone number	EP1067Y
Purification technique	Affinity purification Protein A
Specificity	This antibody detects Vav2 phosphorylated on Tyr172 as well as unphosphorylated Vav2.
Concentration	0.117 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IHC-P

Tested

Species	Human
Dilution info	-
Notes	

	Perform heat-mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

Expected

Species	Mouse
Dilution info	-
Notes	Perform heat-mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

Predicted

Species	Rat
Dilution info	-
Notes	-

IP

Tested

Species	Human
Dilution info	-
Notes	-

Expected

Species	Mouse
Dilution info	Use at an assay dependent concentration.
Notes	-

Predicted

Species	Rat
Dilution info	-
Notes	-

WB

Tested

Species	Human
Dilution info	1/20000
Notes	-

Expected

Species	Mouse
Dilution info	1/20000
Notes	-

Predicted

Species	Rat
Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	1/2500
Notes	-

Expected

Species	Mouse
Dilution info	1/2500
Notes	-

Predicted

Species	Rat
Dilution info	-

Notes	-

Target data

See full target information Guanine nucleotide exchange factor VAV2

Function	Guanine nucleotide exchange factor for the Rho family of Ras-related GTPases. Plays an important role in angiogenesis. Its recruitment by phosphorylated EPHA2 is critical for EFNA1-induced RAC1 GTPase activation and vascular endothelial cell migration and assembly (By similarity).

Storage

Shipped at conditions	Conditional Ambient
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	-20°C
Storage information	Stable for 12 months at -20°C

Notes

Patented technology
Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.

What are the advantages of a recombinant monoclonal antibody?
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 batch production

For more information, read more on recombinant antibodies.

Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

Activity summary	VAV2 also known as Vav 2 guanine nucleotide exchange factor is a member of the VAV family of proteins which function as guanine nucleotide exchange factors (GEFs). It has a molecular mass of approximately 100 kDa. The protein converts

	inactive GDP-bound Rho family GTPases such as Rac1 and Cdc42 into their active GTP-bound form. VAV2 is expressed in various tissues including the brain heart and lung indicating its broad role in cellular signaling across different systems.
Biological function summary	VAV2 influences multiple cellular processes through its role as a GEF. This protein does not form a part of a static complex but interacts dynamically with other signaling components. VAV2 regulates cytoskeleton organization cell migration and growth by activating Rho family GTPases. Its activity impacts processes such as neuronal development and immune response by modifying the actin cytoskeleton which is important for cell movement and morphological changes.
Pathways	VAV2 plays an integral role in signaling cascades like the Ras and MAPK pathways. It interacts closely with proteins like Rac1 and Cdc42 to propagate signals that influence cell proliferation and survival. By facilitating the switch from inactive GDP-bound states to active GTP-bound forms of Rho GTPases VAV2 integrates signals from cell surface receptors to downstream effectors that mediate cellular responses to external stimuli.
Associated diseases and disorders	VAV2’s function connects to cancer progression and cardiovascular disorders. Changes in VAV2 expression or activity can lead to abnormal cell growth and differentiation linking it to cancerous transformations. Additionally because VAV2 modulates cell migration and cytoskeletal dynamics its malfunction can contribute to cardiovascular diseases by affecting vascular smooth muscle cells and endothelial cells. In both contexts VAV2's interactions with Rho family proteins like Rac1 highlights its influence in disease mechanisms.

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

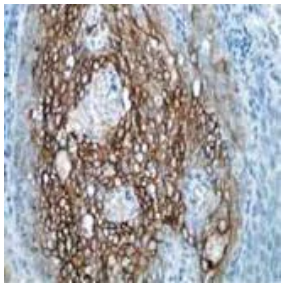
We do not recommend this combination. It is not covered by our product promise.

We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:
Terms & Conditions.

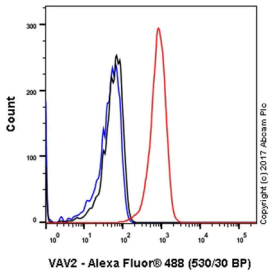
9 product images



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-VAV2 antibody [EP1067Y] (ab52640)

VAV2 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-VAV2 antibody

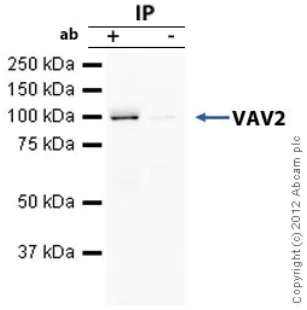
Human cervical carcinoma stained with ab52640 at 1/50 - 1/100 dilution
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Flow Cytometry (Intracellular) - Anti-VAV2 antibody [EP1067Y] (ab52640)

VAV2 Flow Cytometry (Intracellular) staining using rabbit Anti-VAV2 antibody

Intracellular Flow Cytometry analysis of HeLa (human cervix adenocarcinoma) cells labeling VAV2 (red) with ab52640 at a 1/2500 dilution. Cells were fixed with 4% paraformaldehyde and permeabilized with 90% methanol. A goat anti-rabbit IgG (Alexa Fluorr® 488) (ab150077) was used as the secondary antibody at a 1/2000 dilution. Black - Rabbit monoclonal IgG (ab172730). Blue (unlabeled control) - Cells without incubation with the primary and secondary antibodies.

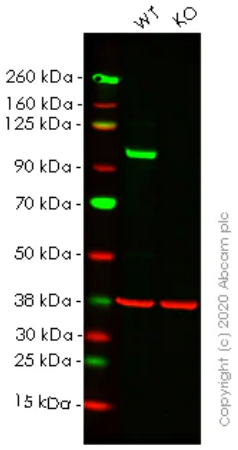


Immunoprecipitation - Anti-VAV2 antibody [EP1067Y] (ab52640)

VAV2 was immunoprecipitated using 0.5mg Hek293 whole cell extract, 10µg of Rabbit monoclonal to VAV2 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, Hek293 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 ab52640.
Secondary: Mouse monoclonal [SB62a] Secondary Antibody to Rabbit IgG light chain (HRP) (ab99697).
Band: 100kDa; VAV2.
All lanes:
Immunoprecipitation - Anti-VAV2 antibody [EP1067Y] (ab52640)

Predicted band size: 101 kDa

Western blot - Anti-VAV2 antibody [EP1067Y] (ab52640)



VAV2 Western blot staining using rabbit Anti-VAV2 antibody

Lanes 1- 2: Merged signal (red and green). Green - ab52640 observed at 100 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (ab8245) observed at 37 kDa.
ab52640 was shown to react with VAV2 in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line ab265318 (knockout cell lysate ab257794) was used. Wild-type HeLa and VAV2 knockout HeLa cell lysates were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. ab52640 and Anti-GAPDH antibody [6C5] - Loading Control (ab8245) overnight at 4°C at a 1 in 20000 dilution and a 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

All lanes:
Western blot - Anti-VAV2 antibody [EP1067Y] (ab52640) at 1/20000 dilution

Lane 1:
Wild-type HeLa cell lysate at 20 µg

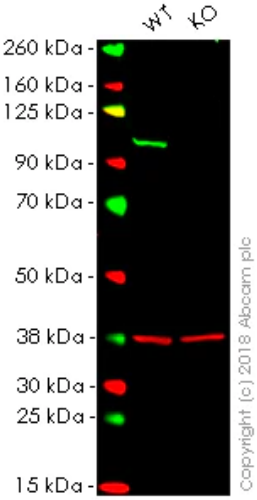
Lane 2:
VAV2 knockout HeLa cell lysate at 20 µg

Performed under reducing conditions.

Predicted band size: 101 kDa

Observed band size: 100 kDa

Western blot - Anti-VAV2 antibody [EP1067Y] (ab52640)



VAV2 Western blot staining using rabbit Anti-VAV2 antibody

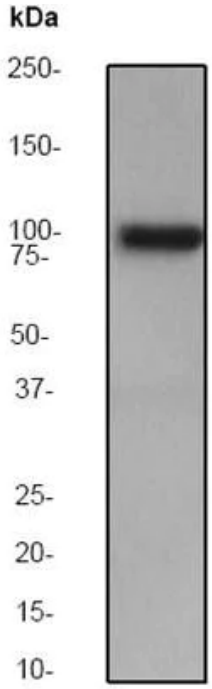
Lanes 1 - 4: Merged signal (red and green). Green - ab52640 observed at 101 kDa. Red - loading control, ab9484, observed at 37 kDa.
ab52640 was shown to specifically react with VAV2 in wild-type HAP1 cells as signal was lost in VAV2 knockout cells. Wild-type and VAV2 knockout samples were subjected to SDS-PAGE. ab52640 and ab9484 (Mouse anti GAPDH loading control) were incubated overnight at 4°C at 1/20000 dilution and 1/20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed ab216773 and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed ab216776 secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.

All lanes:
Western blot - Anti-VAV2 antibody [EP1067Y] (ab52640) at 1/20000 dilution

Lane 1:
Wild-type HAP1 whole cell lysate at 20 µg

Lane 2:
VAV2 knockout HAP1 whole cell lysate at 20 µg

Predicted band size: 101 kDa



Western blot - Anti-VAV2 antibody [EP1067Y] (ab52640)

All lanes:
Western blot - Anti-VAV2 antibody [EP1067Y] (ab52640) at 1/20000 dilution

All lanes:
293 cell lysate at 10 µg

Secondary

All lanes:
Goat anti-rabbit HRP labeled at 1/2000 dilution

Predicted band size: 101 kDa

Observed band size: 101 kDa

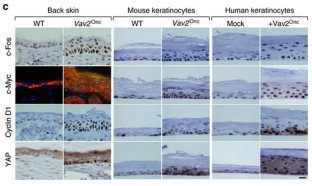


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Immunohistochemistry - Anti-VAV2 antibody [EP1067Y] (ab52640)

VAV2 Immunohistochemistry staining using rabbit Anti-VAV2 antibody

Immunohistochemistry using Anti-VAV2 antibody [EP1067Y], ab52640. Publication image from Lorenzo-Martín, L. F. et al., 2020, Nat Commun, 32963234. Legend direct from paper.

Transcriptional factors involved in the Vav2Onc-dependent transcriptome.a Transcription factor binding sites found enriched in the promoter regions of up- (red) and downregulated (blue) genes present in the “redundant” fraction of the Vav2Onc-driven transcriptome. The NES and percentage of hits for each transcription factor binding site are also indicated. b Circos plot of the differentially expressed genes in the Vav2Onc mouse skin. For each gene, the expression heatmap, the chromosomal location (1–19, X, Y), the binding sites for the indicated transcription factors, and the mirror-image behavior in the Vav2Onc-regulated transcriptome and the Vav2;Vav3-dependent fraction of the TPA-stimulated gene expression program in the skin of mice are indicated. Red, redundant; Nonred, nonreduntant. c Expression of indicated proteins (left) in the epidermis of the back skin of WT and Vav2Onc/Onc mice (left), 3D organotypic cultures from WT and Vav2Onc/Onc primary keratinocytes (middle), and 3D cultures of human keratinocytes (right). In the case of the immunofluorescence experiments using c-Myc antibodies (back skin, green color), sections were decorated with K14 (red color) and DAPI (blue color). Scale bar, 10 µm (n = 3 independent analyses for each experimental group). d Expression of indicated proteins (left) in 3D cultures of control and Rac1F28L + RhoAF30L-expressing human keratinocytes. Scale bar, 10 µm (n = 3 independent cultures). e–h Transiently transfected Vav2Onc triggers the rapid activation of endogenous AP1 (e), c-MYC (f), E2F- (g), and TEAD (h) proteins in human keratinocytes. Activity was measured using luciferase-encoding vectors containing promoter regions for each of the interrogated transcriptional factors, as described in Methods. Data represent the mean ± SEM. Black and gray stars indicate the P value of the indicated experimental values when compared to EGFP- and EGFP-Vav2Onc-transfected cells, respectively. *P = 0.011 (Vav2Onc+E200A) versus Vav2Onc, TEAD); **P = 0.008 (Vav2Onc versus EGFP, AP1), 0.010 (Vav2Onc+E200A) versus Vav2Onc, AP1), 0.002 (Vav2Onc versus EGFP, E2F); ***P < 0.0001 (all other tests) (ANOVA and Tukey’ s HSD test, n = 3 independent experiments). i Expression of indicated proteins in one of the experiments performed in e to h. Tubulin was used as loading control (bottom panel) (n = 3 independent experiments). j,k Correlation between the levels of the VAV2 mRNA and the expression of c-MYC- (j) and YAP/TEAD-regulated- (k) gene signatures in the indicated (top) cSCC (n = 40) and hnSCC (n = 685) gene expression datasets. **P = 0.007 (VAV2 medium, MYC signature, GSE30784), 0.005 (VAV2 high, MYC signature, HPV- TCGA), 0.008 (VAV2 high, YAP signature, GSE45216), 0.002 (VAV2 medium, YAP

signature, GSE30784); ***, P < 0.0001 (all other experiments) using the ANOVA and Dunnett's multiple comparison test). Data represent the mean ± SEM. Source data for this figure are provided as a Source Data file.

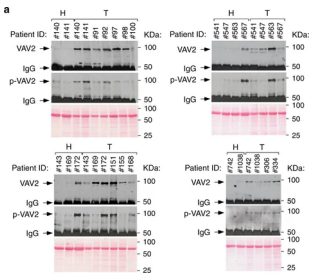


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Western blot - Anti-VAV2 antibody [EP1067Y] (ab52640)

VAV2 Western blot staining using rabbit Anti-VAV2 antibody

Western Blotting using Anti-VAV2 antibody [EP1067Y], ab52640. Publication image from Lorenzo-Martín, L. F. et al., 2020, Nat Commun, 32963234. Legend direct from paper.

VAV2 protein is overexpressed in hnSCC.a Levels of total (VAV2) and tyrosine-phosphorylated (p-VAV2) VAV2 in healthy (H) and tumor (T) tissues from hnSCC patients (n = 83). The band corresponding to the antibody used for the immunoprecipitation step is indicated (IgG). Amount of total protein content in the extracts used for the immunoprecipitation was tested in parallel filters using Ponceau staining (bottom panels). KDa, kilodalton. b VAV2 abundance according to data from a. ***P < 0.0001 (two-tailed Mann–Whitney test, n = 83 patient samples). a.u., arbitrary units. c VAV2 tyrosine phosphorylation levels according to data obtained in a. *P = 0.011 (two-tailed Mann–Whitney test, n = 83 patient samples). Please, note that the total levels of tyrosine phosphorylation of VAV2 in these analyses are probably underestimated given the long experimental procedure associated with tumor collection and subsequent lysis. d Example of tumor sections showing the indicated VAV2 immunoreactivity levels (IRL, top). The percentages of tumor samples (% out of a total of 232 tumors analyzed) showing the indicated VAV2 immunoreactivity levels are shown at the bottom. n = 232 tumor samples. Scale bar, 200 µm. e Representative image showing the lack of VAV2 immunoreactivity in terminally differentiated areas (asterisk) of VAV2-positive tumors. Scale bar, 100 µm. f Representative images of the immunohistochemical staining of VAV2 in healthy (left) and dysplastic (right) samples from human oral epithelium. Sections were counterstained with hematoxylin. n = 232 tumor samples. Scale bar, 100 µm. g Expression levels of the indicated transcripts according to the differentiation status of keratinocytes. Data were obtained from the GEO GSE52651 dataset (see "Methods"). IVL involucrin (an mRNA expressed from the early stages of keratinocyte terminal differentiation); LOR loricrin (a transcript encoding a major protein component of the cornified cell envelope that is found in terminally differentiated epidermal cells). In b and c, data represent the mean ± SEM. Source data for this figure are provided as a Source Data file.

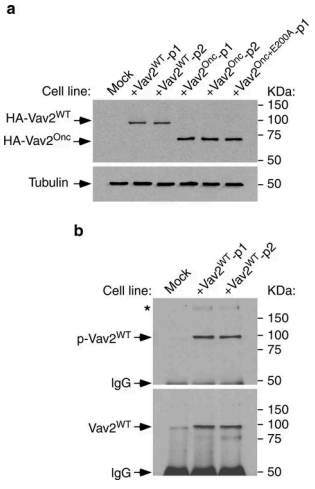


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VAV2 Western blot staining using rabbit Anti-VAV2 antibody

Western Blotting using Anti-VAV2 antibody [EP1067Y], ab52640. Publication image from Lorenzo-Martín, L. F. et al., 2020, Nat Commun, 32963234. Legend direct from paper.

Wild-type VAV2 also triggers keratinocyte hyperplasia.a Immunoblots showing the expression of the indicated Vav2 proteins in human keratinocytes (top panel). Tubulin was used as loading control in all cases (bottom panel) (n = 3 independent experiments). p1 and p2 refer to two independent pools of cells. b Tyrosine-phosphorylation levels (top panel) of immunoprecipitated Vav2WT (bottom panel) in the keratinocyte cell lines. Similar data were obtained in two additional independent experiments. p, tyrosine-phosphorylated. IgG, immunoglobulin band derived from the antibody used in the immunoprecipitation step. The asterisk marks a high molecular weight tyrosine-phosphorylated protein that coimmunoprecipitates with Vav2. c Representative images of hematoxylin/eosin-stained organotypic cultures generated by the indicated cells (top). The thickness of the corneum and cellular epidermal strata is indicated with red and blue brackets, respectively. Scale bar, 10 µm (n = 3 independent cultures). d Thickness of the cellular and corneum strata using data from c. *P = 0.013; ***P < 0.0001 (ANOVA and Dunnett's multiple comparison test, n = 3 independent cultures). Data represent the mean ± SEM. Source data for this figure are provided as a Source Data file.

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