# abcam

#### Product datasheet

## Anti-VAV1 (phospho Y174) antibody [EP510Y] ab76225

Recombinant RabMAb

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Overview

Product name Anti-VAV1 (phospho Y174) antibody [EP510Y]

**Description** Rabbit monoclonal [EP510Y] to VAV1 (phospho Y174)

Host species Rabbit

**Tested applications** Suitable for: Flow Cyt (Intra), WB, ICC/IF, Dot blot

Unsuitable for: IHC-P or IP

Species reactivity Reacts with: Human

**Immunogen** Synthetic peptide within Human VAV1 (phospho Y174). The exact sequence is proprietary.

Positive control WB: Jurkat cell lysate treated with pervanadate. ICC/IF: Jurkat cell lysate treated with

pervanadate. Flow Cyt (intra): WB: Jurkat cell lysate treated with pervanadate.

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<sup>®</sup> technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**<sup>®</sup> **patents**.

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. Upon delivery aliquot and store at -20°C. Avoid freeze / thaw cycles.

Storage buffer pH: 7.20

Preservative: 0.05% Sodium azide

Constituents: 0.1% BSA, 40% Glycerol (glycerin, glycerine), 9.85% Tris glycine, 50% Tissue

culture supernatant

Purity Protein A purified

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Clonality Monoclonal
Clone number EP510Y
Isotype IgG

#### **Applications**

The Abpromise guarantee Our Abpromise guarantee covers the use of ab76225 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
Flow Cyt (Intra)		Use at an assay dependent concentration.
WB	**** (1)	1/1000. Detects a band of approximately 101 kDa (predicted molecular weight: 101 kDa).
ICC/IF		1/100 - 1/500.
Dot blot		Use at an assay dependent concentration.

**Application notes** Is unsuitable for IHC-P or IP.

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Function Couples tyrosine kinase signals with the activation of the Rho/Rac GTPases, thus leading to cell

differentiation and/or proliferation.

**Tissue specificity** Widely expressed in hematopoietic cells but not in other cell types.

Sequence similarities Contains 1 CH (calponin-homology) domain.

Contains 1 DH (DBL-homology) domain.

Contains 1 PH domain.

Contains 1 phorbol-ester/DAG-type zinc finger.

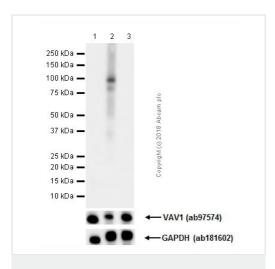
Contains 1 SH2 domain. Contains 2 SH3 domains.

**Domain** The DH domain is involved in interaction with CCPG1.

Post-translational modifications

Phosphorylated on tyrosine residues.

#### **Images**



Western blot - Anti-VAV1 (phospho Y174) antibody [EP510Y] (ab76225)

**All lanes :** Anti-VAV1 (phospho Y174) antibody [EP510Y] (ab76225) at 1/1000 dilution

**Lane 1 :** Jurkat (human T cell leukemia T lymphocyte) whole cell lysate

**Lane 2 :** Jurkat treated with 50mM pervanadate for 5minutes whole cell lysate

Lane 3 : Jurkat treated with 50mM pervanadate for 5min whole cell lysate 10  $\mu g$ . Then the membrane was incubated with alkaline phosphatase

Lysates/proteins at 10 µg per lane.

### Secondary

**All lanes :** Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

Predicted band size: 101 kDa

ab76225 DAPI MERGED

ab76225 DAPI MERGED

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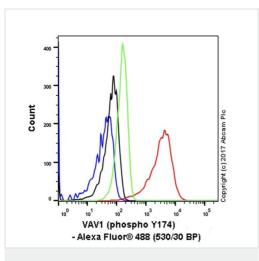
Immunocytochemistry/ Immunofluorescence - Anti-VAV1 (phospho Y174) antibody [EP510Y] (ab76225)

Blocking and diluting buffer: 5% NFDM/TBST

Immunocytochemistry/Immunofluorescence analysis of Untreated Jurkat cells and Pervanadate (1mM,30min) treated Jurkat cells labelling VAV1 (phospho Y174) with purified ab76225 at 1/500. Cells were fixed with 4% PFA and permeabilized with 0.1% Triton X-100, and counterstained with <a href="mailto:ab7291">ab7291</a> anti-Tubulin (mouse mAb) 1:1000 (1ug/ml). An Alexa Fluor<sup>®</sup> 488-conjugated goat anti-rabbit lgG (1/1000) was used as the secondary antibody (Ab150077). Nuclei counterstained with DAPI (blue).

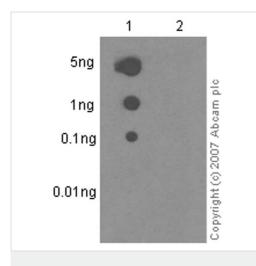
Negative Control 1: Rabbit primary antibody and anti-mouse secondary antibody(<u>ab150120</u>)

Negative Control 2: Mouse primary antibody(<u>ab7291</u>) and antirabbit secondary antibody(<u>ab150077</u>)



Flow Cytometry (Intracellular) - Anti-VAV1 (phospho Y174) antibody [EP510Y] (ab76225)

Intracellular Flow Cytometry analysis of Jurkat (human acute T cell leukemia) untreated/treated with 1mM pervanadate for 30min cells labeling VAV1 with unpurified ab76225 at 1/200 dilution (10ug/ml) (red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. A Goat anti rabbit lgG (Alexa Fluorr<sup>®</sup> 488) (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) were used as the unlabeled control. Untreated control (Green).

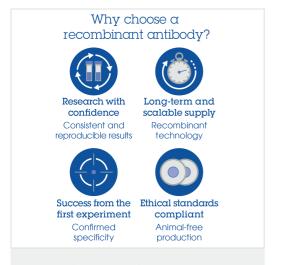


Dot Blot - Anti-VAV1 (phospho Y174) antibody [EP510Y] (ab76225)

Dot blot analysis of VAV1 (pY174) peptide (Lane 1) and VAV1 non-phospho peptide (Lane 2) labelling VAV1 (phospho Y174) with ab76225 at a dilution of 1/1000. A Peroxidase-conjugated goat anti-rabbit lgG (H+L) was used as the secondary antibody at a dilution of 1/2500.

Blocking and dilution buffer: 5% NFDM/TBST.

Exposure time: 3 minutes.



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