abcam

ab217673

# Anti-Vimentin (phospho S56) antibody [EPR21084]

Lab Essentials | Recombinant | RabMAb | 20ul selling size

## Key facts

| | |
|---|---|
| **Isotype** | IgG |
| **Host species** | Rabbit |
| **Storage buffer** | pH: 7.2 - 7.4 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA |
| **Form** | Liquid |
| **Clonality** | Monoclonal |
| **Immunogen** | The exact immunogen used to generate this antibody is proprietary information. |
| **Clone number** | EPR21084 |
| **Purification technique** | Affinity purification Protein A |
| **Concentration** | 0.573 mg/mL The concentration of this product may be batch-dependent<br>Batch concentration finder → |

## Reactivity data

### IP

#### Tested

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 1/30 |
| **Notes** | - |

#### Not recommended

| | |
|---|---|
| **Species** | Synthetic peptide |
| **Dilution info** | - |
| **Notes** | - |

## Dot

### Tested

| | |
|---|---|
| **Species** | Synthetic peptide |
| **Dilution info** | 1/1000 |
| **Notes** | - |

### Expected

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | Use at an assay dependent concentration. |
| **Notes** | - |

## WB

### Tested

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 1/1000 |
| **Notes** | - |

### Not recommended

| | |
|---|---|
| **Species** | Synthetic peptide |
| **Dilution info** | - |
| **Notes** | - |

## ICC/IF

### Tested

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 1/1000 |
| **Notes** | - |

### Not recommended

| | |
|---|---|
| **Species** | Synthetic peptide |
| **Dilution info** | - |
| **Notes** | - |

## Flow Cyt (Intra)

### Tested

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | - |
| **Notes** | - |

### Not recommended

| | |
|---|---|
| **Species** | Synthetic peptide |
| **Dilution info** | - |
| **Notes** | - |

## Target data

See full target information VIM pS56

| | |
|---|---|
| **Function** | Vimentins are class-III intermediate filaments found in various non-epithelial cells, especially mesenchymal cells. Vimentin is attached to the nucleus, endoplasmic reticulum, and mitochondria, either laterally or terminally. Plays a role in cell directional movement, orientation, cell sheet organization and Golgi complex polarization at the cell migration front (By similarity). Protects SCRIB from proteasomal degradation and facilitates its localization to intermediate filaments in a cell contact-mediated manner (By similarity). May promote axon outgrowth and motor fiber repair via DSP-mediated recruitment to outgrowth tips (By similarity). Involved with LARP6 in the stabilization of type I collagen mRNAs for CO1A1 and CO1A2. |

## Storage

| | |
|---|---|
| **Shipped at conditions** | Conditional Ambient |
| **Appropriate short-term storage duration** | 1-2 weeks |
| **Appropriate short-term storage conditions** | +4°C |
| **Appropriate long-term storage conditions** | -20°C |
| **Aliquoting information** | Upon delivery aliquot |
| **Storage information** | Avoid freeze / thaw cycle |

## 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.

## 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**

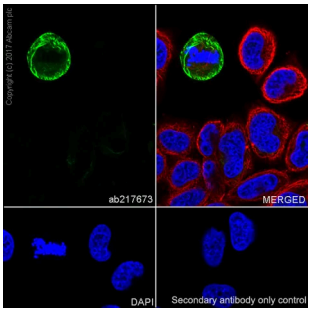
We do not recommend this combination. It is not covered by our product promise.

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.

## 6 product images

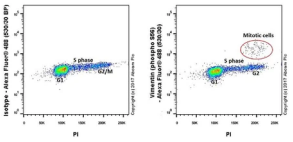



### Immunocytochemistry/ Immunofluorescence - Anti-Vimentin (phospho S56) antibody [EPR21084] (ab217673)

Vimentin (phospho S56) Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-Vimentin (phospho S56) antibody

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (human epithelial cell line from cervix adenocarcinoma) cells labeling Vimentin (phospho S56) with ab217673 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green). Confocal image showing strong positive staining in HeLa cells in M phase.
The nuclear counterstain is DAPI (blue). Tubulin is detected with Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) (ab195889) at 1/200 dilution (red).

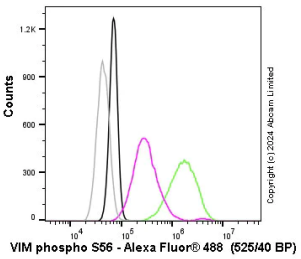
Secondary antibody only control: PBS instead of primary antibody, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution.



### Flow Cytometry (Intracellular) - Anti-Vimentin (phospho S56) antibody [EPR21084] (ab217673)

Vimentin (phospho S56) Flow Cytometry (Intracellular) staining using rabbit Anti-Vimentin (phospho S56) antibody

ab217673 staining Vimentin (phospho S56) in HeLa (human cervix adenocarcinoma epithelial cell) cells by intracellular flow cytometry. Cells were fixed using 80% Methanol and permeabilized with 0.1% Tween-20. The sample was incubated with primary antibody at 1/500 dilution. An Alexa Fluor®488 Goat anti-rabbit IgG (ab150077) was used as a secondary antibody. Rabbit monoclonal IgG (ab172730) was used as an isotype control (left). Cells were pre-treated with 20 µg/ml RNase A for 30 minutes to eliminate the non-specific binding between RNA and PI (Propidium iodide). Vimentiin (phospho S56) is highly expressed in mitotic cells. (PMID: 16260496)




## Flow Cytometry (Intracellular) - Anti-Vimentin (phospho S56) antibody [EPR21084] (ab217673)

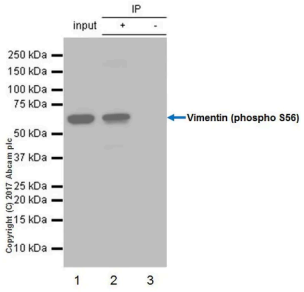
Vimentin (phospho S56) Flow Cytometry (Intracellular) staining of Treated HeLa cells using rabbit Anti-Vimentin (phospho S56) antibody

Flow cytometry overlay histogram showing HeLa treated with 100ng/mL Nocodazole for 18 hours + RNAse (green line) and untreated HeLa (magenta line) stained with ab217673. The cells were fixed with 80% methanol (5 min) and then permeabilised with 0.1% PBS-Tween 20 for 20 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 (ab217673) (1x $10^6$in 100µl at 0.04µg/ml (1/56000)) 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

IIsotype control antibody Recombinant Rabbit IgG, monoclonal [EPR25A] - Isotype Control was used at the same concentration and conditions as the primary antibody (treated HeLa - black line, untreated HeLa - grey line). Unlabelled sample was also used as a control (this line is not shown for the purpose of simplicity)

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




## Immunoprecipitation - Anti-Vimentin (phospho S56) antibody [EPR21084] (ab217673)

Vimentin (phospho S56) was immunoprecipitated from 0.35 mg of HeLa (human epithelial cell line from cervix adenocarcinoma) treated with 100 ng/ml nocodazole (ab120630) for 18 hours, whole cell lysate with ab217673 at 1/30 dilution. Western blot was performed from the immunoprecipitate using ab217673 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/10000 dilution.
**Lane 1:** HeLa treated with 100 ng/ml nocodazole (ab120630) for 18 hours, whole cell lysate 10 µg (Input).
**Lane 2:** ab217673 IP in HeLa treated with 100 ng/ml nocodazole (ab120630) for 18 hours, whole cell lysate.
**Lane 3:** Rabbit monoclonal IgG (ab172730) instead of ab217673 in HeLa treated with 100 ng/ml nocodazole (ab120630) for 18 hours, whole cell lysate.

**Blocking and dilution buffer and concentration:** 5% NFDM/TBST.

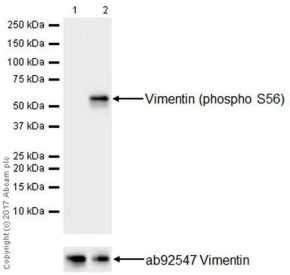
All lanes:
Immunoprecipitation - Anti-Vimentin (phospho S56) antibody [EPR21084] (ab217673)

Developed using the ECL technique.

Predicted band size: 53 kDa

Observed band size: 57 kDa

Exposure time: 1s




## Western blot - Anti-Vimentin (phospho S56) antibody [EPR21084] (ab217673)

Vimentin (phospho S56) Western blot staining using rabbit Anti-Vimentin (phospho S56) antibody

**Blocking/Dilution buffer:** 5% NFDM/TBST.
All lanes:
Western blot - Anti-Vimentin (phospho S56) antibody [EPR21084] (ab217673) at 1/1000 dilution

Lane 1:
Untreated HeLa (human epithelial cell line from cervix adenocarcinoma) whole cell lysate at 10 µg

Lane 2:
HeLa (human epithelial cell line from cervix adenocarcinoma) treated with 100 ng/ml nocodazole (ab120630) for 18 hours, whole cell lysate at 10 µg

Secondary

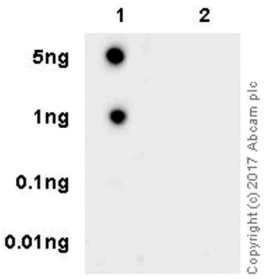
All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Developed using the ECL technique.

Predicted band size: 53 kDa

Observed band size: 57 kDa

Exposure time: 23s




## Dot Blot - Anti-Vimentin (phospho S56) antibody [EPR21084] (ab217673)

Dot blot analysis of Vimentin (phospho S56) labeled with ab217673 at 1/1000 dilution.
**Lane 1:** Vimentin (phospho S56) peptide.
**Lane 2:** Vimentin non-phospho peptide.

Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution was used as secondary antibody.

**Blocking and dilution buffer:** 5% NFDM/TBST.

**Exposure time:** 1 minute.

The blot was developed on a BIO-RAD® ChemiDoc™ MP instrument.

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.