abcam

Product datasheet

Anti-Vimentin antibody [EPR3776] - BSA and Azide free ab193555

KO VALIDATED Recombinant RabMAb

4 Abreviews 48 References 33 Images

Overview

Product name	Anti-Vimentin antibody [EPR3776] - BSA and Azide free
Description	Rabbit monoclonal [EPR3776] to Vimentin - BSA and Azide free
Host species	Rabbit
Tested applications	**Suitable for:** WB, mIHC, Flow Cyt (Intra), IHC-P, ICC/IF
Species reactivity	**Reacts with:** Mouse, Rat, Human, African green monkey **Predicted to work with:** Rhesus monkey
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: HeLa, HEK293, Jurkat, A549, NIH3T3, PC12, HUVEC, Daudi, Caco-2 and COS-1 cell lysates; mouse and rat brain tissue lysates. IHC-P: Human kidney, colon, breast adenocarcinoma, cervical carcinoma and ovarian cancer tissues, mouse brain and kidney, E17 rat cheek and rat skin tissue sections; Rhesus monkey retina tissue. IHC-Fr: Mouse testis tissue. ICC/IF: HeLa, human adenocarcinoma, human schlemms canal endothelium and wild-type HAP1 cells. Flow Cyt (intra): HeLa cells. mIHC: Human testis
General notes	ab193555 is the carrier-free version of **ab92547**. Our **carrier-free** antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency. This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications. Use our **conjugation kits** for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold. This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc. This product is a recombinant monoclonal antibody, which offers several advantages including: - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity

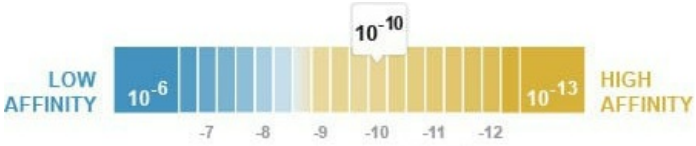
- Long-term security of supply
- Animal-free production

For more information **see here**.

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

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C. Do Not Freeze.
Dissociation constant (K_D)	K_D = 1.10 x 10^{-10} M



Learn more about K_D

Storage buffer	pH: 7.2 Constituent: PBS
Carrier free	Yes
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR3776
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab193555 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

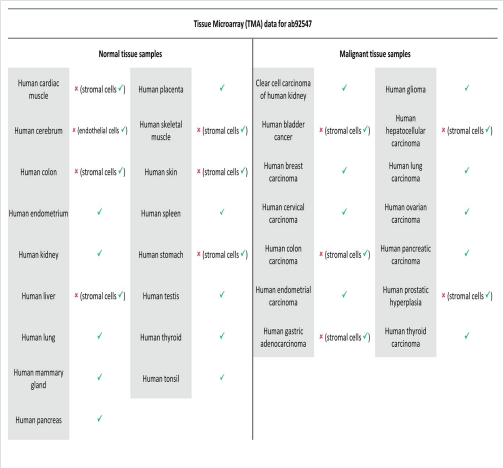
Application	Abreviews	Notes
WB		Use at an assay dependent concentration. Predicted molecular weight: 54 kDa.
mIHC		Use at an assay dependent concentration.
Flow Cyt (Intra)		Use at an assay dependent concentration. **ab199376** - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
IHC-P	★★★★★ **(2)**	Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Application	Abreviews	Notes
ICC/IF		Use at an assay dependent concentration. This product gave a positive signal in HeLa (VIM knockout HeLa cells were used as a negative control) fixed with 4% formaldehyde (10 min) and 100% methanol (5 min).

Target

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. Involved with LARP6 in the stabilization of type I collagen mRNAs for CO1A1 and CO1A2.
Tissue specificity	Highly expressed in fibroblasts, some expression in T- and B-lymphocytes, and little or no expression in Burkitt's lymphoma cell lines. Expressed in many hormone-independent mammary carcinoma cell lines.
Involvement in disease	Cataract 30
Sequence similarities	Belongs to the intermediate filament family.
Domain	The central alpha-helical coiled-coil rod region mediates elementary homodimerization. The [IL]-x-C-x-x-[DE] motif is a proposed target motif for cysteine S-nitrosylation mediated by the iNOS-S100A8/A9 transnitrosylase complex.
Post-translational modifications	Filament disassembly during mitosis is promoted by phosphorylation at Ser-55 as well as by nestin (By similarity). One of the most prominent phosphoproteins in various cells of mesenchymal origin. Phosphorylation is enhanced during cell division, at which time vimentin filaments are significantly reorganized. Phosphorylation by PKN1 inhibits the formation of filaments. Phosphorylated at Ser-56 by CDK5 during neutrophil secretion in the cytoplasm. Phosphorylated by STK33. O-glycosylated during cytokinesis at sites identical or close to phosphorylation sites, this interferes with the phosphorylation status. S-nitrosylation is induced by interferon-gamma and oxidatively-modified low-densitity lipoprotein (LDL(ox)) possibly implicating the iNOS-S100A8/9 transnitrosylase complex.
Cellular localization	Cytoplasm.
Form	Vimentin is found in connective tissue and in the cytoskeleton.

Images

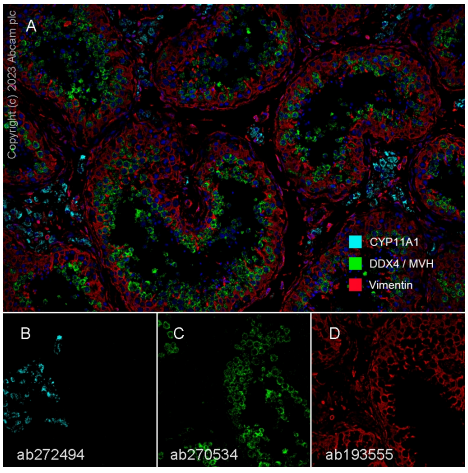


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

This data was developed using **ab92547**, the same antibody clone in a different buffer formulation.

Tissue Microarrays stained for Anti-Vimentin antibody [EPR3776] - Cytoskeleton Marker using **ab92547** in immunohistochemical analysis. This table provides a detailed overview of positive (tick mark) and negaive (cross mark) staining per sample type tested. The section was incubated with **ab92547** at 4°C overnight followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP polymer) secondary antibody (**ab214880**).

Heat mediated antigen retrieval was performed using **ab93684** (Tris/EDTA buffer, pH 9.0).



Multiplex immunohistochemistry - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

Multiplex immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human testis tissue.

Panel A: Merged staining of anti-Vimentin (ab193555; red; Opal™690), anti-CYP11A1 (**ab272494**; cyan; Opal™520) and anti-DDX4 / MVH (**ab270534**; green; Opal™570) on human testis.

Panel B: Anti-CYP11A1 stained on Leydig cells.

Panel C: Anti-DDX4 / MVH stained on all spermatogenic cell types.

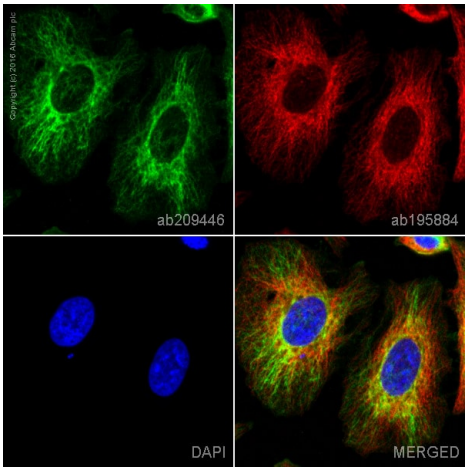
Panel D: Anti-Vimentin stained on Sertoli cells and fibroblasts.

Key protocol steps: The section was incubated in three rounds of staining: in the order of ab193555 (1:2000 dilution) and **ab272494** (1:10000 dilution) for 30 mins, then **ab270534** (1:2000 dlilution) for 10 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system.

The immunostaining was performed on a Leica Biosystems BOND® RX instrument with an Opal™ 4-color kit. Image acquisition was performed with Leica SP8 confocal microscope.

DAPI was used as a nuclear counter stain. Opal Polymer HRP Ms + Rb was used as a secondary.

Antigen retrieval: Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins



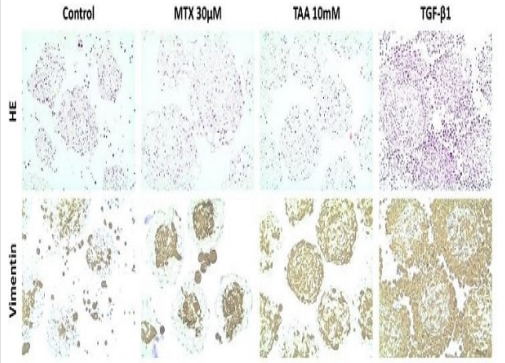
Immunocytochemistry/ Immunofluorescence - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

Clone EPR3776 (ab193555) has been successfully conjugated by Abcam. This image was generated using Anti-Vimentin antibody [EPR3776] - Cytoskeleton Marker (PE). Please refer to **ab209446** for protocol details.

ab209446 staining Vimentin in HeLa cells. The cells were fixed with 100% methanol (5min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated overnight at +4°C with **ab209446** at 1/500 dilution (Pseudocolored in green) and **ab195884**, Rat monoclonal to Tubulin (Alexa Fluor® 647), at 1/250 dilution (shown in red). Nuclear DNA was labelled with DAPI (shown in blue).

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

This product also gave a positive signal under the same testing conditions in HeLa cells fixed with 4% formaldehyde (10 min).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

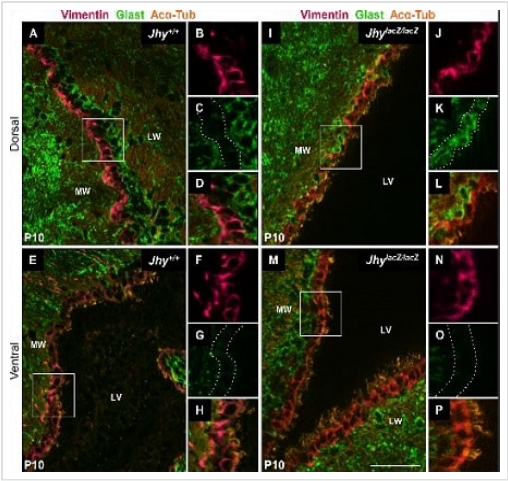
Image from Prestigiacomo V et al. PLoS One. 2017;12(6):e0179995. Fig 7.; doi: 10.1371/journal.pone.0179995.

Immunostaining of formalin fixed paraffin embedded human microtissues after exposure to MTX, TAA and TGF-β1.

Formalin fixed paraffin embedded slides of HepaRG/THP-1 macrophages/hTERT-HSC microtissues were stained with Hematoxylin & Eosin (H&E) and vimentin after 14 days of treatment with MTX, TAA and TGF-β1. Microtissues were fixed in 4% PFA and embedded in 2% agarose prior to paraffinization. Microtissues showed increase in the vimentin positive cells after MTX, TAA and TGF-β1 exposure. Vimentin stainings show proliferation of stellate cells and THP-1 macrophages in the microtissues, suggesting the onset of inflammation process.

For full image see PMID 28665955.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab92547**).



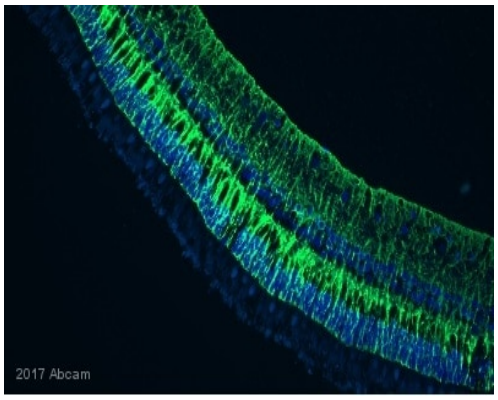
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

Image from Muniz-Talavera H and Schmidt JV. PLoS One. 2017;12(12):e0184957. Fig 3.; doi: 10.1371/journal.pone.0184957.

Jhy$^{lacZ/lacZ}$ mice exhibit delayed radial glial to ependymal cell differentiation.

Immunohistochemical analysis of P10 lateral ventricle coronal sections from *Jhy*$^{+/+}$ (A, E) and *Jhy*$^{lacZ/lacZ}$ (I, M) mice for expression of Vimentin (pink, **ab92547**), Glast (green) and Acα-Tub (orange) in dorsal (A-D, I-L) and ventral (E-H, M-P) brain regions. Lower right panels (D, L, H, P) represent a higher magnification view of the merged image. In *Jhy*$^{+/+}$, medial wall dorsal and ventral cells express the differentiated ependymal markers Vimentin (A, B, E, F) and Acα-Tub (A, D, E, H), but are negative for the radial glial marker Glast (A, C, E, G). In *Jhy*$^{lacZ/lacZ}$ brains, some dorsal cells remain positive for the undifferentiated marker Glast (I, K), while also expressing the differentiated markers Vimentin and Acα-Tub (I, J, L). *Jhy*$^{lacZ/lacZ}$ ventral cells express only Vimentin and Acα-Tub (M-P). The dotted line indicates the medial wall ependymal cells in (C, G, K, O). (Q-R) Graphical representation of the percentage of Glast(-)Vimentin(+)Acα-Tub(+) (black bar) and Glast(+)Vimentin(+)Acα-Tub(+) (grey bar) cells in dorsal (Q) and ventral (R) ependymal cells. MW, medial wall; LW, lateral wall; LV, lateral ventricle; * denotes p≤0.05. Scale bars: 50μm (A-P).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab92547**).

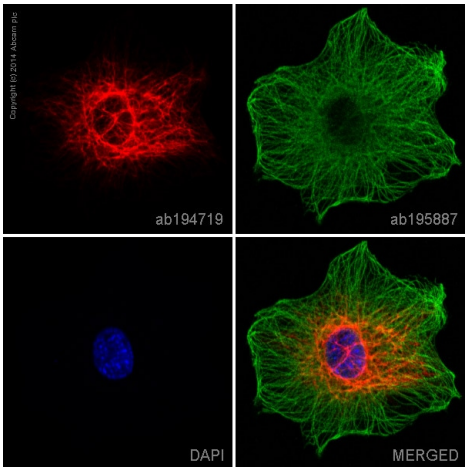


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

Formalin-fixed, paraffin-embedded rhesus monkey retina tissue stained for Vimentin (green) using **ab92547** at 1/200 dilution in ICC/IF.

Goat anti-rabbit AlexaFluor 488 was used as the secondary.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab92547**).

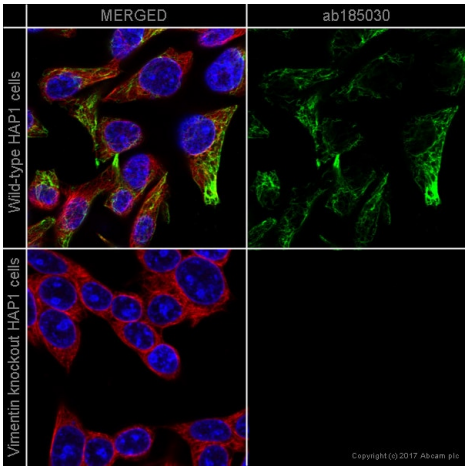


Immunocytochemistry/ Immunofluorescence - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

Clone EPR3776 (ab193555) has been successfully conjugated by Abcam. This image was generated using Anti-Vimentin antibody [EPR3776] - Cytoskeleton Marker (Alexa Fluor® 647). Please refer to **ab194719** for protocol details.

ab194719 staining Vimentin in NIH3T3 cells. The cells were fixed with 4% formaldehyde (10 min), permeabilized in 0.1% Triton X-100 for 5 minutes and then blocked in 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with **ab194719** at 1/100 dilution(shown in red) and **ab195887**, Mouse monoclonal [DM1A] to alpha Tubulin (Alexa Fluor® 488, shown in green) at 2µg/ml overnight at +4°C. Nuclear DNA was labelled in blue with DAPI.

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

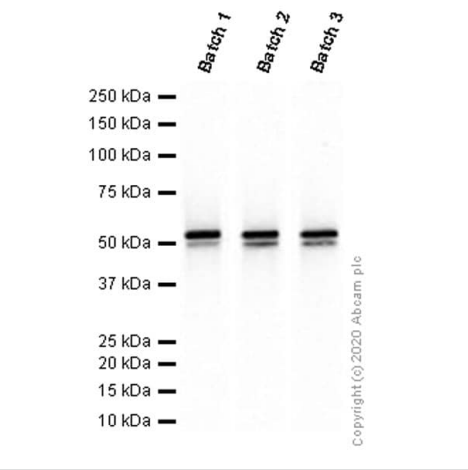


Immunocytochemistry/ Immunofluorescence - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

Clone EPR3776 (ab193555) has been successfully conjugated by Abcam. This image was generated using Anti-Vimentin antibody [EPR3776] - Cytoskeleton Marker (Alexa Fluor® 488). Please refer to **ab185030** for protocol details.

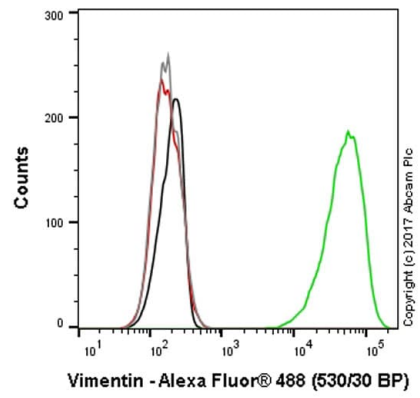
ab185030 staining Vimentin in wild-type HAP1 cells (top panel) and Vimentin knockout HAP1 cells (bottom panel). The cells were fixed with 100% methanol (5min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated overnight at +4°C with **ab185030** at a 1/500 dilution (shown in green) and **ab195889**, Mouse monoclonal to alpha Tubulin (Alexa Fluor® 594), at a 1/250 dilution (shown in red). Nuclear DNA was labelled with DAPI (shown in blue).

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).



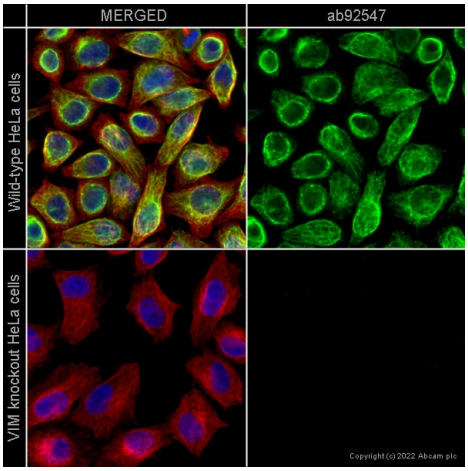
Western blot - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

This data was developed using **ab92547**, the same antibody clone in a different buffer formulation. Different batches of **ab92547** were tested on HEK-293 (Human embryonic kidney epithelial cell) lysate at 0.02 µg/ml. 15 µg of lysate was loaded in each lane. Bands observed at 54 kDa.



Flow Cytometry (Intracellular) - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

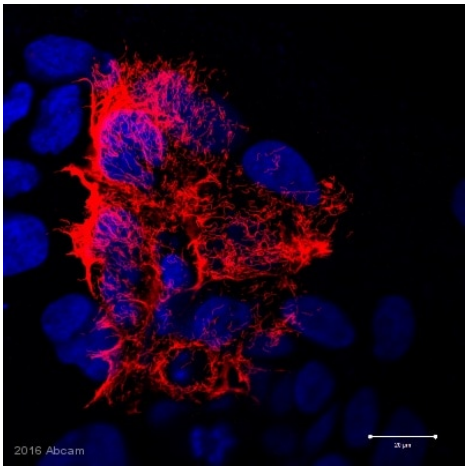
Overlay histogram showing HAP1 wildtype (green line) and HAP1-VIM knockout cells (red line) stained with **ab92547**. The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Triton X-100 for 15 min. The cells were then incubated in 1x PBS / 10% normal goat serum to block non-specific protein-protein interactions followed by the antibody (**ab92547**, 0.5µg/ml) for 30 min at 22°C. The secondary antibody used was Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed (**ab150081**) secondary antibody at 1/2000 dilution for 30 min at 22°C. A Rabbit IgG isotype control antibody (**ab172730**) was used at the same concentration and conditions as the primary antibody (HAP1 wildtype - black line, HAP1-VIM knockout - grey line). Unlabelled sample was also used as a control (this line is not shown for the purpose of simplicity). Acquisition of >5,000 events were collected using a 50 mW Blue laser (488nm) and 530/30 bandpass filter. This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab92547**).



Immunocytochemistry/ Immunofluorescence - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

ab92547 staining VIM in wild-type HeLa cells, with negative expression in VIM knockout HeLa cells. The cells were fixed with 4% formaldehyde (10 min), permeabilised with 0.1% Triton x-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated overnight at +4°C with **ab92547** at 2 µg/ml and **ab7291**, Mouse monoclonal [DM1A] to alpha Tubulin at 0.5 µg/ml. Cells were then incubated with **ab150081**, Goat polyclonal Secondary Antibody to Rabbit IgG - H&L (Alexa Fluor® 488), pre-adsorbed at 1/1000 dilution (shown in green) and **ab150119**, Goat polyclonal Secondary Antibody to Mouse IgG - H&L (Alexa Fluor® 647), pre-adsorbed at 1/1000 dilution (shown in red). Nuclear DNA was labelled with DAPI (shown in blue).Image was acquired with a confocal microscope (Leica-Microsystems TCS SP8) and a single confocal section is shown..This product also work with 100% methanol (5 min) fixation under the same testing conditions.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab92547**).

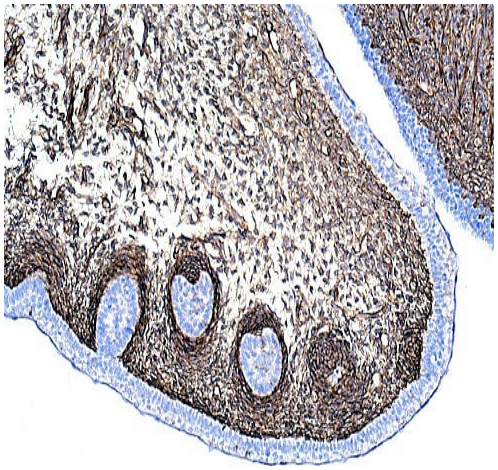


Immunocytochemistry/ Immunofluorescence - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

This image is courtesy of an anonymous Abreview.

ab92547 staining Vimentin in human adenocarcinoma cells by ICC/IF (Immunocytochemistry/immunofluorescence). Cells were fixed with paraformaldehyde, permeabilized with 0.1% TX-100 in PBS and blocked with 5% serum for 1 at 21°C. Samples were incubated with primary antibody (1/400) for 12 hours at 21°C. A CY3® conjugated donkey anti-rabbit polyclonal was used as the secondary antibody at 1/200.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab92547**).

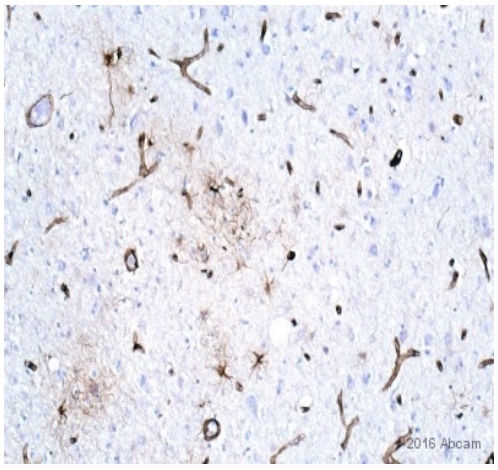


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

Anti-vimentin (**ab92547**) staining in E17 rat cheek sections using immunohistochemistry (formaldehyde-fixed, paraffin-embedded sections). Heat-mediated antigen retrieval was carried out using citric acid. Samples were incubated with primary antibody (1/2000) for two hours at room temperature. A biotin-conjugated goat anti-rabbit IgG polyclonal was used as the secondary antibody.

Image courtesy of Mr Carl Hobbs, Kings College London.

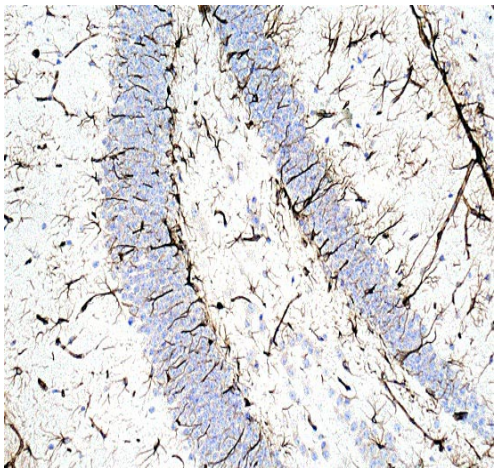
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab92547**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

Formalin-fixed, paraffin-embedded human AD brain tissue stained for vimentin using **ab92547** at 1/2000 dilution in immunohistochemical analysis.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab92547**).

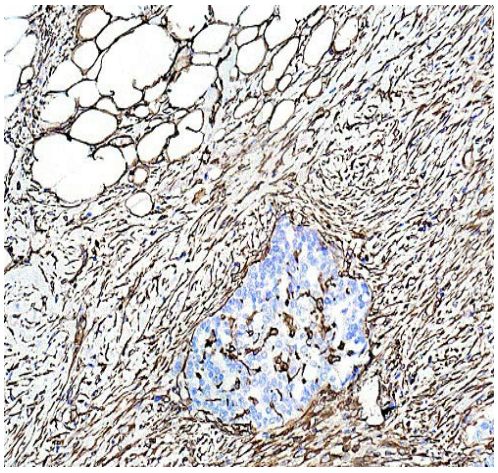


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

Anti-vimentin (**ab92547**) staining in adult mouse brain (the dentate gyrus region of the hippocampus) using immunohistochemistry (formaldehyde-fixed, paraffin-embedded sections). Heat-mediated antigen retrieval was carried out using citric acid. Samples were incubated with primary antibody (1/2000) for two hours at room temperature. A biotin-conjugated goat anti-rabbit IgG polyclonal was used as the secondary antibody.

Image courtesy of Mr Carl Hobbs, Kings College London.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab92547**).

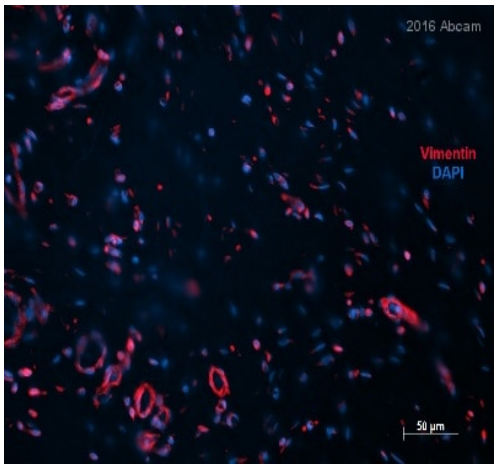


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

Anti-vimentin (**ab92547**) staining in human ovarian cancer tissue using immunohistochemistry (formaldehyde-fixed, paraffin-embedded sections). Heat-mediated antigen retrieval was carried out using citric acid. Samples were incubated with primary antibody (1/2000) for two hours at room temperature. A biotin-conjugated goat anti-rabbit IgG polyclonal was used as the secondary antibody.

Image courtesy of Mr Carl Hobbs, Kings College London.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab92547**).

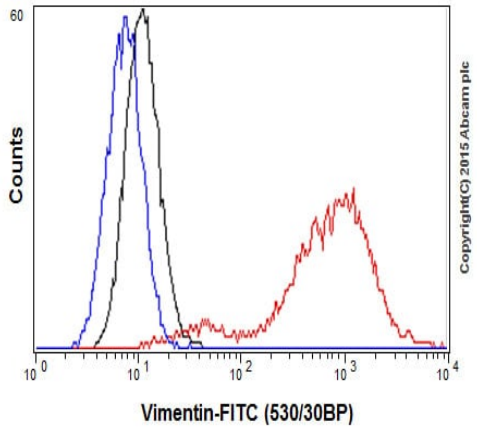


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

This image is courtesy of an anonymous Abreview.

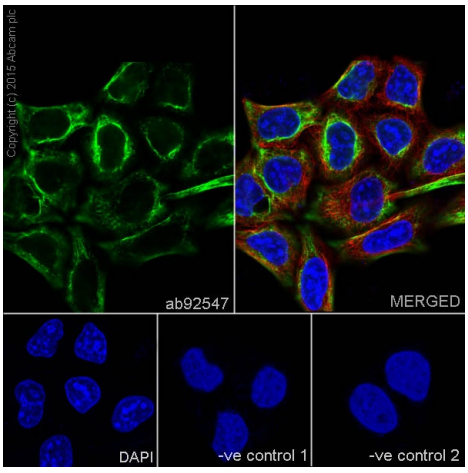
ab92547 staining Vimentin in rat skin tissue sections by Immunohistochemistry (IHC-P - paraformaldehyde-fixed, paraffin-embedded sections). Tissue was fixed with 10% buffered normal formalin and blocked with 5% serum for 60 minutes at 21°C; antigen retrieval was by heat mediation in a 10mM Sodium citrate buffer. Samples were incubated with primary antibody (1/200 in blocking buffer) for 12 hours at 4°C. A Cy3®-conjugated donkey anti-rabbit IgG polyclonal (1/200) was used as the secondary antibody.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab92547**).



Flow Cytometry (Intracellular) - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

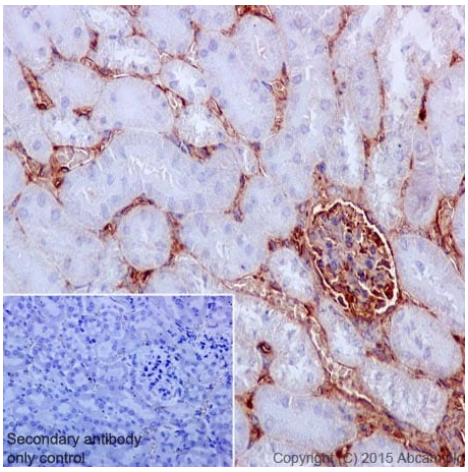
Overlay histogram showing HeLa cells fixed in 2% PFA and stained with purified **ab92547** at a dilution of 1 in 50 (red line). The secondary antibody used was FITC goat anti-rabbit at a dilution of 1 in 500. Rabbit monoclonal IgG was used as an isotype control (black line) and cells incubated in the absence of both primary and secondary antibody were used as a negative control (blue line). This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab92547**).



Immunocytochemistry/ Immunofluorescence - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

Immunofluorescence staining of HeLa (human epithelial cell line from cervix adenocarcinoma) cells with purified **ab92547** at a working dilution of 1/250, counter-stained with DAPI. The secondary antibody was **Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077) secondary antibody**, used at a dilution of 1/1000. **ab7291**, a mouse anti-tubulin antibody (1/1000), was used to stain tubulin along with **Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) preadsorbed (ab150120)** 1/1000, shown in the top right hand panel. The cells were fixed in 4% PFA and permeabilized using 0.1% Triton X 100. The negative controls are shown in bottom middle and right hand panels - for negative control 1, purified **ab92547** was used at a dilution of 1/500 followed by **Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) preadsorbed (ab150120)** at a dilution of 1/500. For negative control 2, **ab7291** (mouse anti-tubulin) was used at a dilution of 1/500 followed by **Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077) secondary antibody** at a dilution of 1/400.

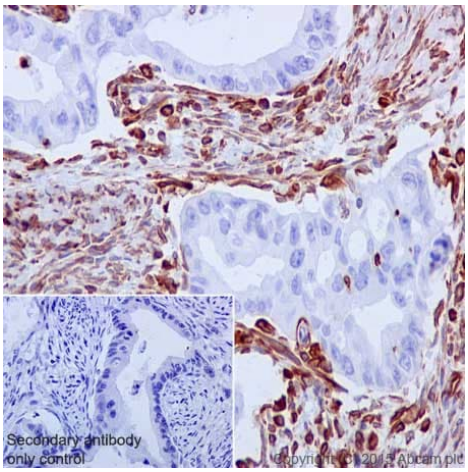
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab92547**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

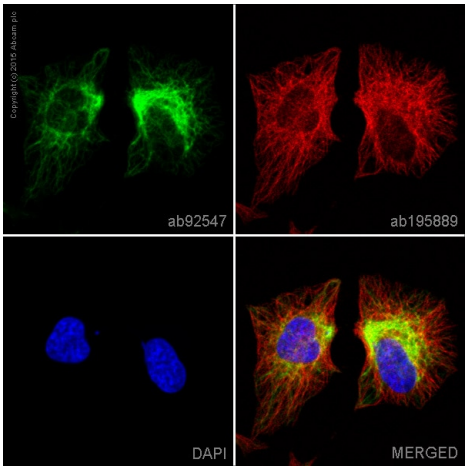
Immunohistochemical staining of paraffin embedded mouse kidney with purified **ab92547** at a working dilution of 1/250. The secondary antibody used is **Goat Anti-Rabbit IgG H&L (HRP) (ab97051) secondary antibody** at 1/500. The sample is counter-stained with hematoxylin. Antigen retrieval was perfomed using Tris-EDTA buffer, pH 9.0. PBS was used instead of the primary antibody as the negative control, and is shown in the inset.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab92547**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

Immunohistochemical staining of paraffin embedded human cervical carcinoma with purified **ab92547** at a working dilution of 1/250. The secondary antibody used is HRP goat anti-rabbit IgG H&L (**ab97051**) at 1/500. The sample is counter-stained with hematoxylin. Antigen retrieval was perfomed using Tris-EDTA buffer, pH 9.0. PBS was used instead of the primary antibody as the negative control, and is shown in the inset.
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab92547**).

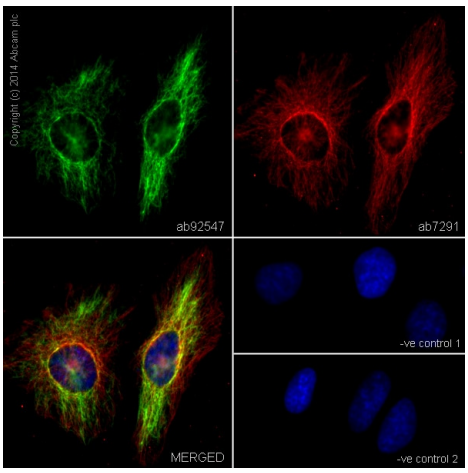


Immunocytochemistry/ Immunofluorescence - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

Unpurified **ab92547** staining Vimentin in HeLa cells. The cells were fixed with 100% methanol (5 min), permeabilized in 0.1% Triton X-100 for 5 minutes and then blocked in 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with **ab92547** at a working concentration of 5µg/ml and **ab195889**, Mouse monoclonal [DM1A] to alpha Tubulin (Alexa Fluor® 594, shown in red) at 1/250 overnight at +4°C, followed by a further incubation at room temperature for 1h with **Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed (ab150081) secondary antibody** at 2 µg/ml (shown in green). Nuclear DNA was labelled in blue with DAPI.

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab92547**).

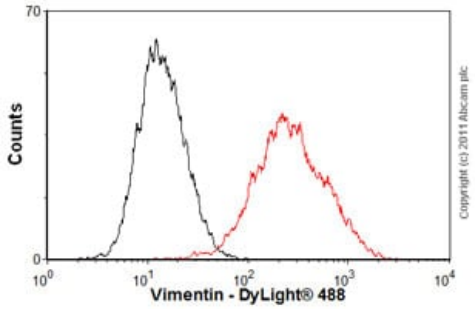


Immunocytochemistry/ Immunofluorescence - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

ab92547 staining Vimentin in HeLa (human epithelial cell line from cervix adenocarcinoma) cells. The cells were fixed with 100% methanol (5min) and then blocked in 1% BSA/10% normal goat serum/0.3M glycine in 0.1%PBS-Tween for 1h. The cells were then incubated with **ab92547** at 5µg/ml and **ab7291** at 1µg/ml overnight at +4°C, followed by a further incubation at room temperature for 1h with **Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed (ab150081) secondary antibody** at 2 µg/ml (shown in green) and **Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) preadsorbed (ab150120)** at 2 µg/ml (shown in pseudo color red). Nuclear DNA was labelled in blue with DAPI.

Negative controls: 1– Rabbit primary antibody and anti-mouse secondary antibody; 2 – Mouse primary antibody and anti-rabbit secondary antibody. Controls 1 and 2 indicate that there is no unspecific reaction between primary and secondary antibodies used.

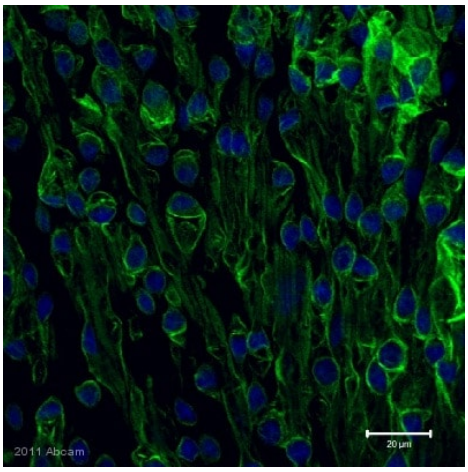
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab92547**).



Flow Cytometry (Intracellular) - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

Overlay histogram showing HeLa cells stained with unpurified **ab92547** (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Triton X-100 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 (**ab92547** , 1/100 dilution) for 30 min at 22°C. The secondary antibody used was Goat Anti-Rabbit IgG H&L (DyLight® 488) preadsorbed (**ab96899**) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) (1µg/1x10^6 cells) used under the same conditions. Acquisition of >5,000 events was performed. This antibody gave a positive signal in HeLa cells fixed with 4% paraformaldehyde/permeabilized in 0.1% PBS-Triton X-100 used under the same conditions.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab92547**).

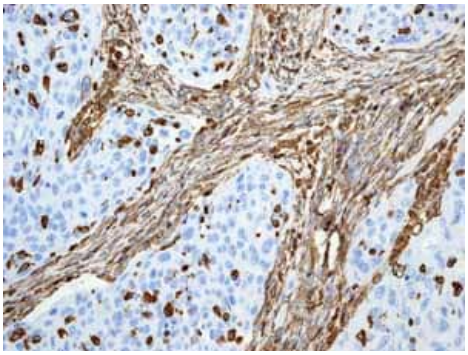


Immunocytochemistry/ Immunofluorescence - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

This image is courtesy of an Abreview submitted by Thomas Read.

Unpurified **ab92547** staining vimentin in human Schlemms Canal Endothelium cells by ICC/IF (Immunocytochemistry/immunofluorescence). Cells were fixed with formaldehyde, permeabilized with Triton X-100 0.2% and blocked with 10% serum for 30 minutes at 20°C. Samples were incubated with primary antibody (1/200 in DPBS) for 3 hours at 20°C. An undiluted Alexa Fluor®488-conjugated Goat anti-rabbit IgG polyclonal was used as the secondary antibody.

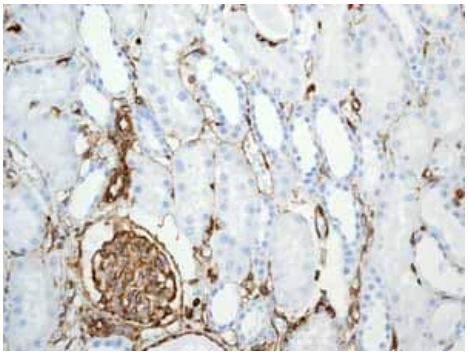
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab92547**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

Immunohistochemical analysis of formalin/PFA-fixed paraffin-embedded human cervical carcinoma tissue sections labeling Vimentin with **ab92547**.

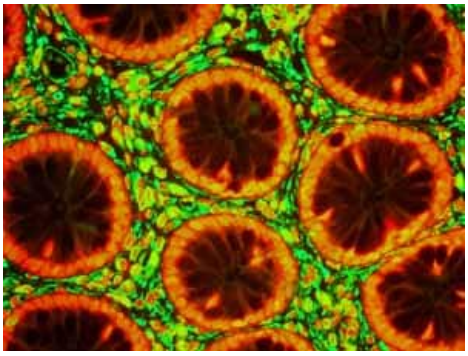
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab92547**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

Immunohistochemical analysis of formalin/PFA-fixed paraffin-embedded human kidney tissue sections labeling Vimentin with **ab92547**.

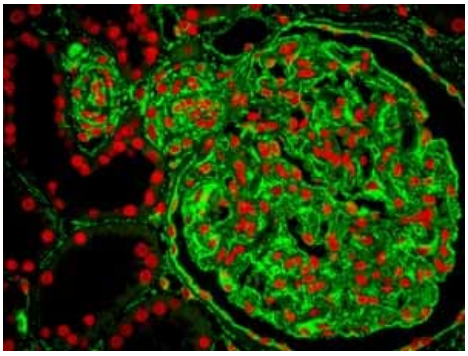
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab92547**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

Fluorescent immunohistochemical analysis of paraffin-embedded human normal colon tissue using unpurified **ab92547**. Green-Vimentin red-PI

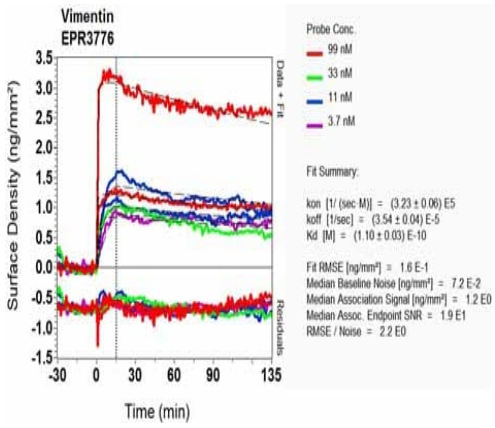
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab92547**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

Fluorescent immunohistochemical analysis of paraffin-embedded human normal kidney tissue using unpurified **ab92547**. Green-Vimentin red-PI.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab92547**).



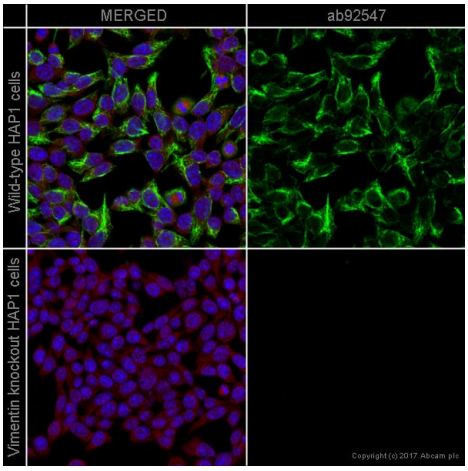
OI-RD Scanning - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

Equilibrium disassociation constant (K_D)

Learn more about K_D

Click here to learn more about K_D

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab92547**).

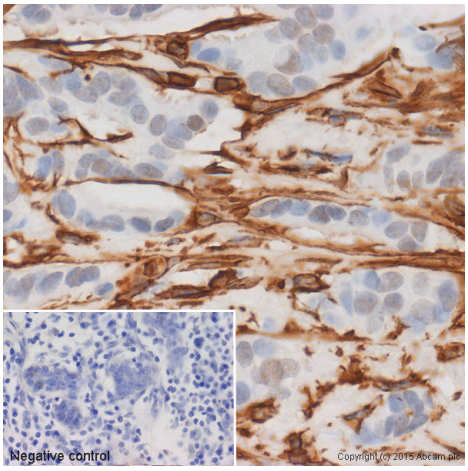


Immunocytochemistry/ Immunofluorescence - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

This ICC data was generated using the same anti-Vimentin antibody clone, EPR3776, in a different buffer formulation (cat# **ab92547**).

ab92547 staining Vimentin in wild-type HAP1 cells (top panel) and VIM knockout HAP1 cells (bottom panel). The cells were fixed with 100% methanol (5min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with **ab92547** at 0.5µg/ml and **ab195889** at 1/250 dilution (shown in pseudocolour red) overnight at +4°C, followed by a further incubation at room temperature for 1h with a goat secondary antibody to Rabbit IgG (Alexa Fluor® 488) (**ab150081**) at 2 µg/ml (shown in green). Nuclear DNA was labelled in blue with DAPI.

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).



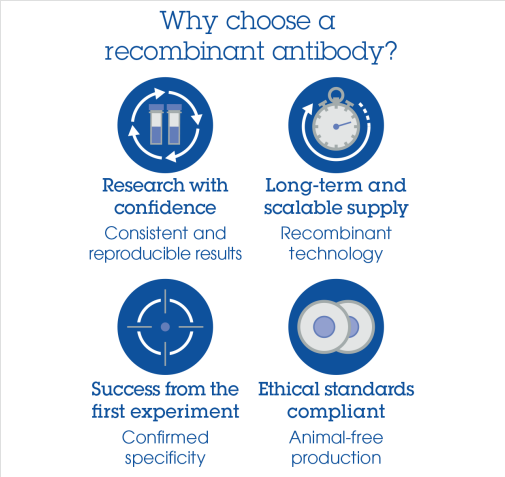
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

This IHC data was generated using the same anti-Vimentin antibody clone, EPR3776, in a different buffer formulation (cat# **ab92547**).

IHC image of unpurified **ab92547** staining Vimentin in human breast adenocarcinoma formalin-fixed paraffin-embedded tissue sections*, performed on a Leica Bond. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with **ab92547**, 1/200 dilution, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX. No primary antibody was used in the negative control (shown on the inset).

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.

*Tissue obtained from the Human Research Tissue Bank, supported by the NIHR Cambridge Biomedical Research Centre



Anti-Vimentin antibody [EPR3776] - BSA and Azide free (ab193555)

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