abcam

Product datasheet

Anti-alpha Tubulin antibody [EP1332Y] - BSA and Azide free ab216650

Recombinant RabMAb

21 References 13 Images

Overview

Product name	Anti-alpha Tubulin antibody [EP1332Y] - BSA and Azide free
Description	Rabbit monoclonal [EP1332Y] to alpha Tubulin - BSA and Azide free
Host species	Rabbit
Specificity	This antibody is expected to recognise most alpha tubulin proteins and not only TUBA4A.
Tested applications	**Suitable for:** Flow Cyt (Intra), WB, IHC-P, ICC/IF
Species reactivity	**Reacts with:** Mouse, Rat, Human, Pig, Drosophila melanogaster
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB- HeLa, HEK-293, HepG2, Caco2, NIH/3T3, PC-12, RAW 264.7, PC-12, C6 Jurkat and HEK-293T whole cell lysates; human fetal kidney lysate; Mouse and rat brain lysate; Pig skeletal muscle lysates; IHC-P: Pig kidney tissue; rat kidney tissue; mouse kidney tissue; human breast cancer and stomach tissue; IHC-Fr: Rat kidney tubule tissue; Flow Cyt (intra): HepG2 cells; ICC/IF: HUVEC, HeLa and 293 cells.
General notes	ab216650 is the carrier-free version of **ab52866**. 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.
Storage buffer	pH: 7.20 Constituent: PBS
Carrier free	Yes
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EP1332Y
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab216650 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. **ab199376**- Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
WB		Use at an assay dependent concentration. Detects a band of approximately 50 kDa (predicted molecular weight: 50 kDa).
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
ICC/IF		Use at an assay dependent concentration.

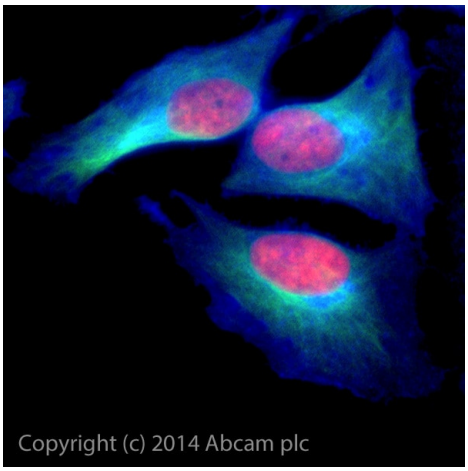
Target

Function	Tubulin is the major constituent of microtubules. It binds two moles of GTP, one at an exchangeable site on the beta chain and one at a non-exchangeable site on the alpha chain.
Sequence similarities	Belongs to the tubulin family.
Post-translational modifications	Some glutamate residues at the C-terminus are polyglutamylated. This modification occurs exclusively on glutamate residues and results in polyglutamate chains on the gamma-carboxyl group. Also monoglycylated but not polyglycylated due to the absence of functional TTLL10 in

human. Monoglycylation is mainly limited to tubulin incorporated into axonemes (cilia and flagella) whereas glutamylation is prevalent in neuronal cells, centrioles, axonemes, and the mitotic spindle. Both modifications can coexist on the same protein on adjacent residues, and lowering glycylation levels increases polyglutamylation, and reciprocally. The precise function of such modifications is still unclear but they regulate the assembly and dynamics of axonemal microtubules.
Acetylation of alpha chains at Lys-40 stabilizes microtubules and affects affinity and processivity of microtubule motors. This modification has a role in multiple cellular functions, ranging from cell motility, cell cycle progression or cell differentiation to intracellular trafficking and signaling.

Cellular localization Cytoplasm > cytoskeleton.

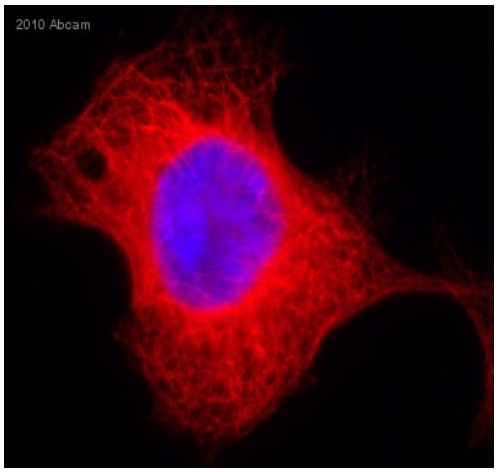
Images



Immunocytochemistry/ Immunofluorescence - Anti-alpha Tubulin antibody [EP1332Y] - BSA and Azide free (ab216650)

Clone EP1332Y (ab216650) has been successfully conjugated by Abcam. This image was generated using Anti-alpha Tubulin antibody [EP1332Y] - Microtubule Marker (Alexa Fluor® 488). Please refer to **ab185031** for protocol details.

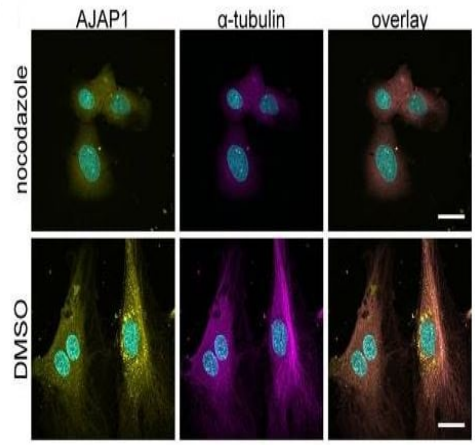
ab185031 staining alpha-Tubulin in HeLa cells. The cells were fixed with 100% methanol (5min) and then blocked in 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with **ab185031** at a working dilution of 1 in 100 overnight at +4°C (shown in green). Alexa Fluor® 350 WGA was used at a 1/200 dilution and incubated for 1h with the cells, to label plasma membranes (shown in blue). Nuclear DNA was labelled in red with 1.25 µM DRAQ5™ (**ab108410**).



Immunocytochemistry/ Immunofluorescence - Anti-alpha Tubulin antibody [EP1332Y] - BSA and Azide free (ab216650)

This image is courtesy of an anonymous Abreview.

ab52866 staining alpha Tubulin in 293 Human embryonic kidney cells by ICC/IF (Immunocytochemistry/immunofluorescence). Cells were fixed with paraformaldehyde and blocked with 10% serum for 2 hours at 23°C. Samples were incubated with primary antibody (1/200 in 0.5% saponin) for 2 hours at 23°C. An Alexa Fluor®555-conjugated Goat anti-rabbit IgG polyclonal (1/500) was used as the secondary antibody. Nuclei were counterstained with DAPI.
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab52866**).



Immunocytochemistry/ Immunofluorescence - Anti-alpha Tubulin antibody [EP1332Y] - BSA and Azide free (ab216650)

Image from Hotte K. et al Biol Open. 2017 Jun 15;6(6):723-731. doi: 10.1242/bio.022335.

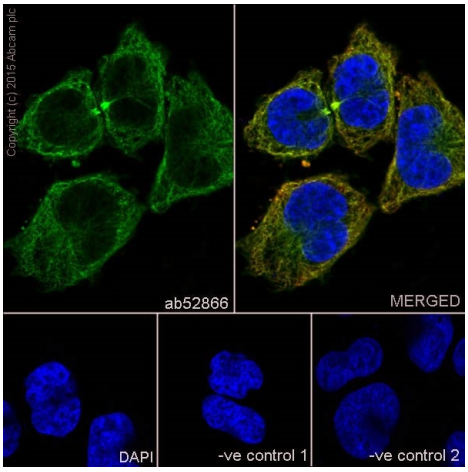
AJAP1 co-localizes with microtubules in HUVECs

The association of AJAP1 with microtubules in HUVECs is lost upon microtubule destruction. Treatment with 12.5 µM nocodazole for 24 h shows destruction of the microtubule network and loss of AJAP1 tubular localization. For a negative control, HUVECs are treated with DMSO for 24 h. Cell nuclei were counterstained with DAPI (cyan). Microscope: Zeiss LSM 780; objective lens: 63×/1.40 oil; scale bar: 25 µm.

Incubated overnight at 4°C with **ab52866**.

(From Figure 3E of Hotte et al)

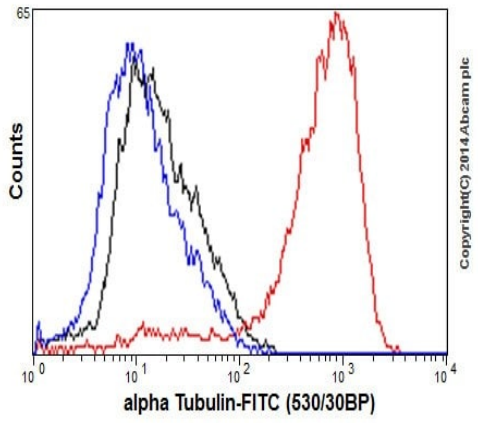
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab52866**).



Immunocytochemistry/ Immunofluorescence - Anti-alpha Tubulin antibody [EP1332Y] - BSA and Azide free (ab216650)

Immunofluorescent analysis of 4% paraformaldehyde-fixed HeLa (human epithelial cell line from cervix adenocarcinoma) cells labeling alpha Tubulin with **ab52866** at 1/500 dilution. The cells were permeabilised with 0.1% Triton X-100. Anti-rabbit Alexa Fluor® 488 (**ab150077**) at 1/400 dilution was used as the secondary antibody (green). The confocal image shows microtubules staining on HeLa cell line. The nuclear counter stain is DAPI (blue). Tubulin is detected with **ab7291** (anti-Tubulin mouse mAb) at 1/500 and anti-mouse AlexaFluor® 594 (**ab150120**) at 1/500 dilution (red).
The negative controls are as follows:
1. **ab52866** at 1/500 dilution followed by anti-mouse AlexaFluor® 594 (**ab150120**) at 1/500 dilution.
2. **ab7291** (anti-Tubulin mouse mAb) at 1/500 dilution followed by anti-rabbit Alexa Fluor® 488 (**ab150077**) at 1/400 dilution.

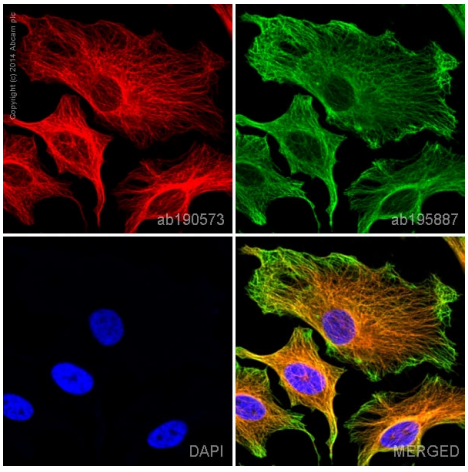
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab52866**).



Flow Cytometry (Intracellular) - Anti-alpha Tubulin antibody [EP1332Y] - BSA and Azide free (ab216650)

Intracellular Flow Cytometry analysis of 2% paraformaldehyde fixed HepG2 (human liver hepatocellular carcinoma cell line) cells labeling alpha Tubulin with **ab52866** at 1/130 dilution (red line). Secondary antibody used is a goat anti rabbit IgG (FITC) at 1/150 dilution. The isotype control is rabbit monoclonal IgG (black line). The unlabeled control is cells without incubation with primary and secondary antibodies (blue line).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab52866**).



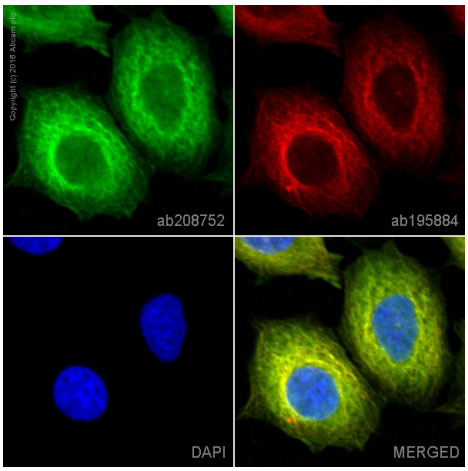
Immunocytochemistry/ Immunofluorescence - Anti-alpha Tubulin antibody [EP1332Y] - BSA and Azide free (ab216650)

Clone EP1332Y (ab216650) has been successfully conjugated by Abcam. This image was generated using Anti-alpha Tubulin antibody [EP1332Y] - Microtubule Marker (Alexa Fluor® 647). Please refer to **ab190573** for protocol details.

ab190573 staining alpha Tubulin in HeLa 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 **ab190573** at a working dilution of 1 in 100 (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.

This product also gave a positive signal in 100% methanol (5 min) fixed HeLa cells under the same testing conditions.

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

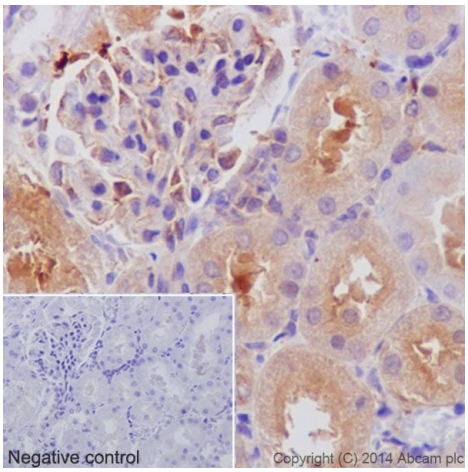


Immunocytochemistry/ Immunofluorescence - Anti-alpha Tubulin antibody [EP1332Y] - BSA and Azide free (ab216650)

Clone EP1332Y (ab216650) has been successfully conjugated by Abcam. This image was generated using Anti-alpha Tubulin antibody [EP1332Y] - Microtubule Marker (PE). Please refer to **ab208752** for protocol details.

ab208752 staining alpha Tubulin in MCF7 cells. The cells were fixed with 4% formaldehyde (10 min), 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 **ab208752** 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).



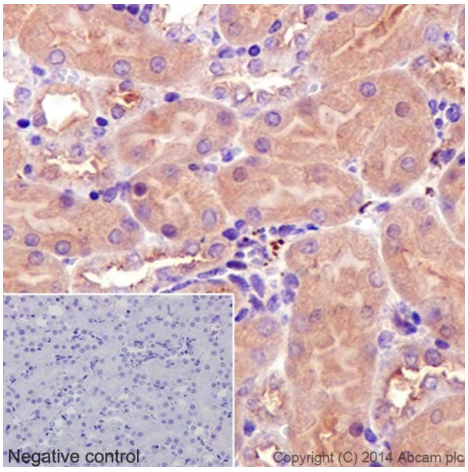
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-alpha Tubulin antibody [EP1332Y] - BSA and Azide free (ab216650)

Immunohistochemistry analysis of paraffin-embedded Rat kidney tissue labeling alpha Tubulin with **ab52866** at a 1/1000 dilution. Cytoplasmic staining on Rat kidney tubule and weak on glomerulus shown. Secondary antibody **Anti-Rabbit HRP** (**ab97051**) used at a 1/500 dilution. Counter stained with Hematoxylin.

Inset image: negative control obtained using PBS instead of **ab52866**, secondary antibody is **Anti-Rabbit HRP** (**ab97051**) at 1/500 dilution.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab52866**).

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



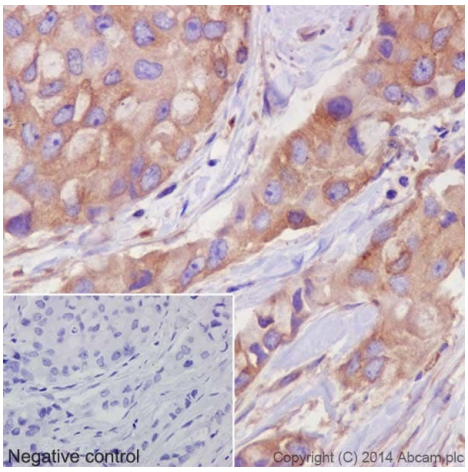
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-alpha Tubulin antibody [EP1332Y] - BSA and Azide free (ab216650)

Immunohistochemistry analysis of paraffin-embedded Mouse kidney tissue labeling alpha Tubulin with **ab52866** at a 1/1000 dilution. Cytoplasmic staining on Mouse kidney tubule shown. Secondary antibody **Anti-Rabbit HRP** (**ab97051**) used at a 1/500 dilution. Counter stained with Hematoxylin.

Inset image: negative control obtained using PBS instead of **ab52866**, secondary antibody is **Anti-Rabbit HRP** (**ab97051**) at 1/500 dilution.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab52866**).

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



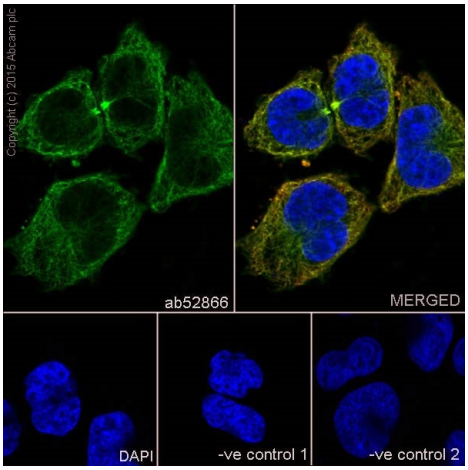
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-alpha Tubulin antibody [EP1332Y] - BSA and Azide free (ab216650)

Immunohistochemistry analysis of paraffin-embedded Human breast cancer labeling alpha Tubulin with **ab52866** at a 1/1000 dilution. Cytoplasmic staining on cancer cells shown. Secondary antibody **ab97051** Goat Anti-Rabbit IgG H&L (HRP) used at a 1/500 dilution. Counter stained with Hematoxylin.

Inset image: negative control obtained using PBS instead of **ab52866**, secondary antibody is Anti-Rabbit HRP (**ab97051**) at 1/500 dilution.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab52866**).

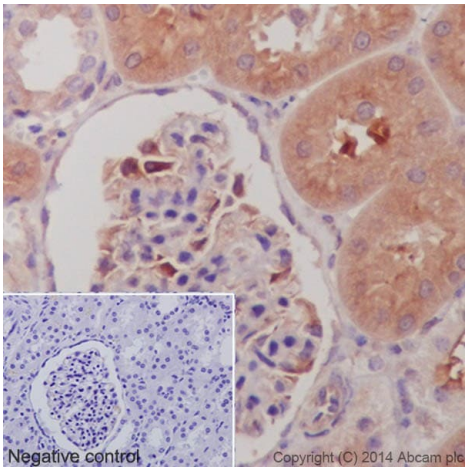
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Immunocytochemistry/ Immunofluorescence - Anti-alpha Tubulin antibody [EP1332Y] - BSA and Azide free (ab216650)

This ICC data was generated using the same anti-alpha Tubulin antibody clone, EP1332Y, in a different buffer formulation (**ab52866**).

Immunofluorescent analysis of 4% paraformaldehyde-fixed HeLa cells labeling alpha Tubulin with **ab52866** at 1/500 dilution. The cells were permeabilised with 0.1% Triton X-100. Anti-rabbit Alexa Fluor® 488 (**ab150077**) at 1/400 dilution was used as the secondary antibody (green). The confocal image shows microtubules staining on HeLa cell line. The nuclear counter stain is DAPI (blue). Tubulin is detected with **ab7291** (anti-Tubulin mouse mAb) at 1/500 and anti-mouse AlexaFluor® 594 (**ab150120**) at 1/500 dilution (red).
The negative controls are as follows:
1. **ab52866** at 1/500 dilution followed by anti-mouse AlexaFluor® 594 (**ab150120**) at 1/500 dilution.
2. **ab7291** (anti-Tubulin mouse mAb) at 1/500 dilution followed by anti-rabbit Alexa Fluor® 488 (**ab150077**) at 1/400 dilution.



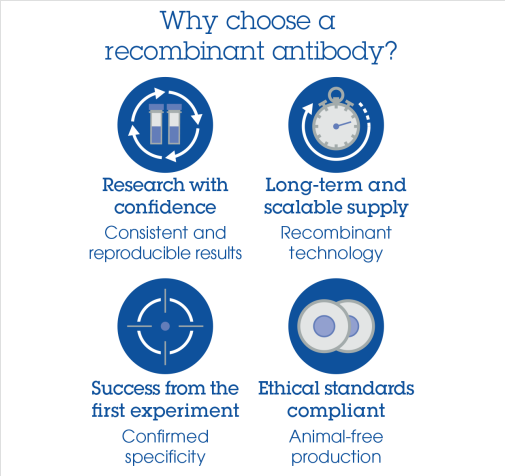
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-alpha Tubulin antibody [EP1332Y] - BSA and Azide free (ab216650)

This IHC data was generated using the same anti-alpha Tubulin antibody clone, EP1332Y, in a different buffer formulation (cat# **ab52866**).

Immunohistochemistry analysis of paraffin-embedded Pig kidney tissue labeling alpha Tubulin with **ab52866** at a 1/1000 dilution. Cytoplasmic staining on Pig kidney tubule and weak on glomerulus shown. **Anti-Rabbit HRP** (**ab97051**) used at a 1/100 dilution. Counter stained with Hematoxylin.

Inset image: negative control obtained using PBS instead of **ab52866**, secondary antibody is **Anti-Rabbit HRP** (**ab97051**) at 1/100 dilution.

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



Anti-alpha Tubulin antibody [EP1332Y] - BSA and Azide free (ab216650)

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