abcam

Product datasheet

Anti-alpha Tubulin antibody [TU-01] ab7750

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Overview

Product name	Anti-alpha Tubulin antibody [TU-01]
Description	Mouse monoclonal [TU-01] to alpha Tubulin
Host species	Mouse
Tested applications	**Suitable for:** Flow Cyt (Intra), IHC-P, ICC/IF, WB
Species reactivity	**Reacts with:** Mouse, Human **Predicted to work with:** Turkey, Pig, Saccharomyces cerevisiae, Paramecium tetraurelia, Nicotiana benthamiana
Immunogen	Full length native protein (purified) corresponding to Pig alpha Tubulin aa 37-154.
Epitope	aa 65-97 on N-terminal structural domain
Positive control	ICC/IF: HeLa and NIH/3T3 cells. IHC-P: Human skin tissue. Flow Cyt (Intra): HEK293 cells, HeLa cells. WB: Jurkat, HeLa, HEK293T, U87-MG all under reducing conditions.
General notes	This product was changed from ascites to tissue culture supernatant on 24th January 2018. Please note that the dilutions may need to be adjusted accordingly. If you have any questions, please do not hesitate to contact our scientific support team. The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets your needs before purchasing. If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, along with publications, customer reviews and Q&As

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C. Do Not Freeze.
Storage buffer	pH: 7.40 Preservative: 0.097% Sodium azide Constituent: PBS
Purity	Proprietary Purification

Purification notes	Purified from TCS. Purified by precipitation and chromatography. Purity >95% by SDS-PAGE.
Clonality	Monoclonal
Clone number	TU-01
Isotype	IgG1

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab7750 in the following tested applications.

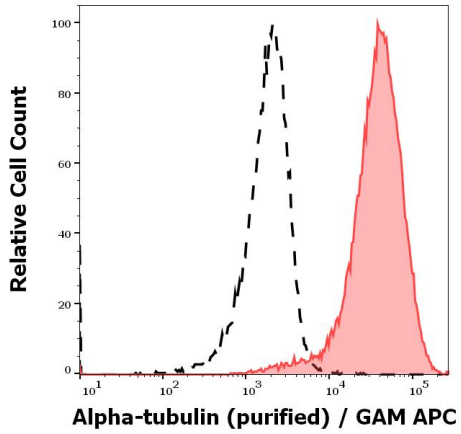
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
Flow Cyt (Intra)		Use a concentration of 1 - 4 µg/ml. **ab170190** - Mouse monoclonal IgG1, is suitable for use as an isotype control with this antibody.
IHC-P		Use a concentration of 2 µg/ml. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.
ICC/IF	★★★☆☆ (1)	Use at an assay dependent concentration.
WB	★★★★☆ (5)	Use a concentration of 1 - 2 µg/ml. Predicted molecular weight: 50 kDa. reducing conditions.

Target

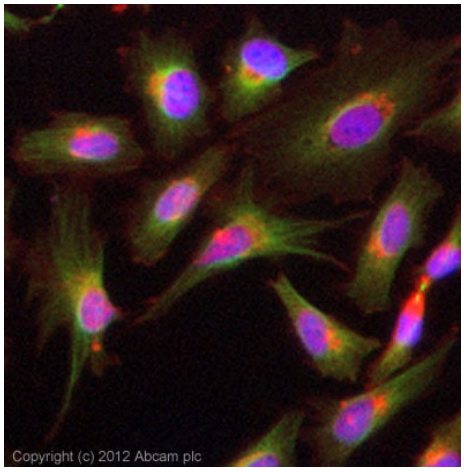
Function	Tubulin is the major constituent of microtubules. It binds two moles of GTP, one at an exchangeable site on the beta chain and one at a non-exchangeable site on the alpha chain.
Sequence similarities	Belongs to the tubulin family.
Post-translational modifications	Some glutamate residues at the 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



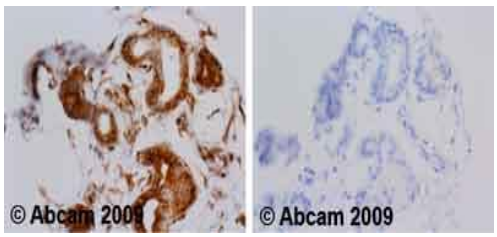
Flow Cytometry (Intracellular) - Anti-alpha Tubulin antibody [TU-01] (ab7750)

Separation of HeLa cells stained using ab7750 (concentration in sample 3 µg/ml, GAM APC, red-filled) from HeLa cells unstained by primary antibody (GAM APC, black-dashed) in flow cytometry analysis (intracellular staining).



Immunocytochemistry/ Immunofluorescence - Anti-alpha Tubulin antibody [TU-01] (ab7750)

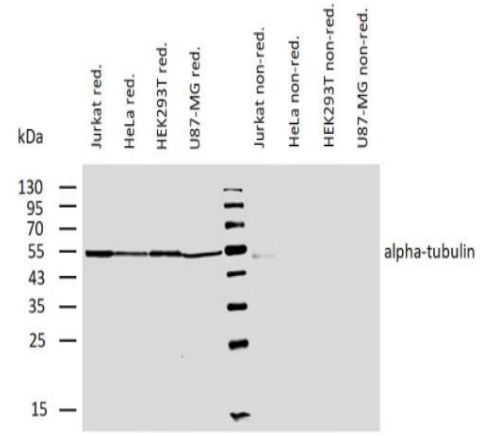
ICC/IF image of ab7750 stained HeLa cells. The cells were 4% formaldehyde fixed (10 min) and then incubated in 1%BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody (ab7750, 5µg/ml) overnight at +4°C. The secondary antibody (green) was **anti-mouse DyLight® 488** (**ab96879**) used at a 1/250 dilution for 1h. Alexa Fluor® 594 WGA was used to label plasma membranes (red) at a 1/200 dilution for 1h. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43µM.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-alpha Tubulin antibody [TU-01] (ab7750)

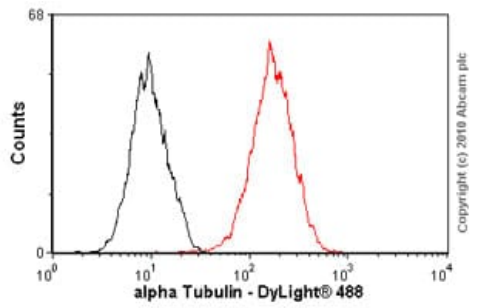
Ab7750 staining human normal skin. Staining is localised to the cytoplasm.
Left panel: with primary antibody at 2 ug/ml. Right panel: isotype control.
Sections were stained using an automated system at room temperature. Sections were rehydrated and antigen retrieved with a retrieval buffer EDTA pH 9.0 . Slides were peroxidase blocked in 3% H_2O_2 in methanol for 10 minutes. They were then blocked for 10 minutes (containing casein 0.25% in PBS) then incubated with primary antibody for 20 minutes and detected with an amplification kit for 30 minutes. Colorimetric detection was completed with diaminobenzidine for 5 minutes. Slides were counterstained with Haematoxylin. Please note that for manual staining we recommend

to optimize the primary antibody concentration and incubation time (overnight incubation), and amplification may be required.



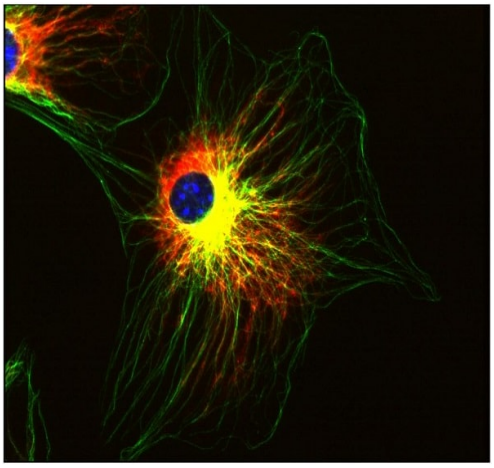
Western blot - Anti-alpha Tubulin antibody [TU-01] (ab7750)

Western blotting analysis labeling alpha-tubulin using ab7750 on lysates of various cell lines under reducing and non-reducing conditions. Nitrocellulose membrane was probed with 2 µg/ml of **ab74696** followed by IRDye800-conjugated streptavidin. A specific band was detected for alpha-tubulin at approximately 54 kDa.



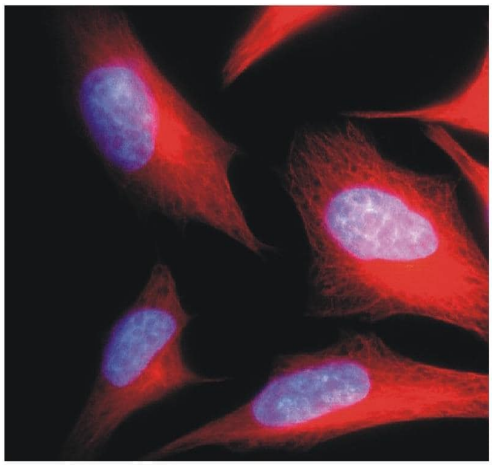
Flow Cytometry (Intracellular) - Anti-alpha Tubulin antibody [TU-01] (ab7750)

Overlay histogram showing HEK293 cells stained with ab7750 (red line). The cells were fixed with 4% paraformaldehyde (10 min) and then permeabilized with 0.1% PBS-Triton 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 (ab7750, 1µg/1x10^6 cells) for 30 min at 22°C. The secondary antibody used was anti-mouse DyLight® 488 (**ab96879**) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was mouse IgG1 [ICIGG1] (**ab91353**, 2µg/1x10^6 cells) used under the same conditions. Acquisition of >5,000 events was performed. This antibody gave a positive signal in HEK293 cells fixed with methanol (5 min)/permeabilized in 0.1% PBS-Triton used under the same conditions.



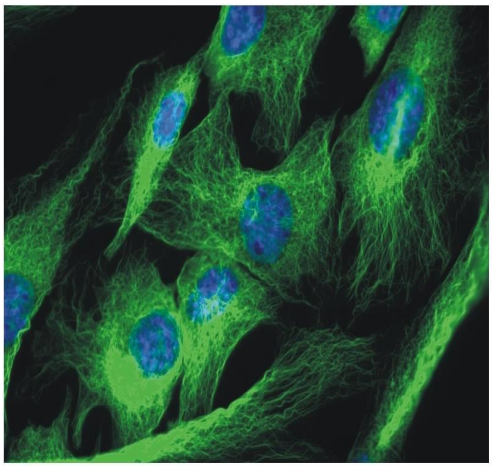
Immunocytochemistry/ Immunofluorescence - Anti-alpha Tubulin antibody [TU-01] (ab7750)

Immunocytochemistry/ Immunofluorescence analysis of NIH 3T3 (mouse embryonal fibroblast) cells labelling alpha tubulin (green) with ab7750. Vimentin was stained red as counterstain. DAPI was used to stain the nuclei blue.



Immunocytochemistry/ Immunofluorescence - Anti-alpha Tubulin antibody [TU-01] (ab7750)

Immunocytochemistry analysis of HeLa (human cervix carcinoma) cells labelling alpha tubulin (red) with ab7750. DAPI was used to stain the nuclei blue.



Immunocytochemistry/ Immunofluorescence - Anti-alpha Tubulin antibody [TU-01] (ab7750)

Immunocytochemistry analysis of NIH 3T3 (mouse embryonal fibroblast) cells labelling alpha tubulin (green) with ab7750. DAPI was used to stain the nuclei blue.

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