abcam

Product datasheet

Anti-TPX2 antibody [18D5-1] ab32795

★★★★★ 3 Abreviews 13 References 7 Images

Overview

Product name	Anti-TPX2 antibody [18D5-1]
Description	Mouse monoclonal [18D5-1] to TPX2
Host species	Mouse
Specificity	This antibody detects TPX2.
Tested applications	**Suitable for:** ICC/IF, IP, WB, IHC-P
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	Recombinant TPX2 protein (Human).

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Upon delivery aliquot and store at -20°C. Avoid freeze / thaw cycles.
Storage buffer	Preservative: 0.05% Sodium Azide Constituents: PBS, 1mg/ml BSA
Purity	Protein G purified
Clonality	Monoclonal
Clone number	18D5-1
Isotype	IgG

Applications

Our Abpromise guarantee covers the use of **ab32795** in the following tested applications.

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

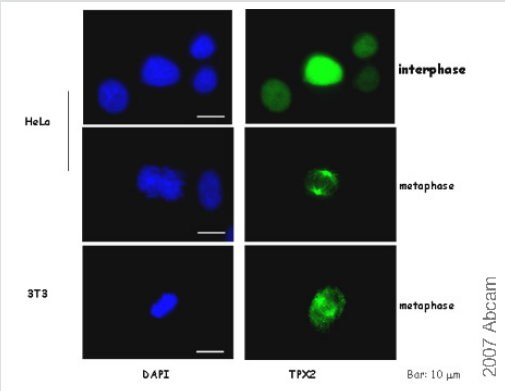
Application	Abreviews	Notes
ICC/IF	★★★★★	Use at an assay dependent dilution. PubMed: 19910498
IP		Use at an assay dependent dilution.

Application	Abreviews	Notes
WB		Use a concentration of 2 µg/ml. Detects a band of approximately 92 kDa (predicted molecular weight: 86 kDa).
IHC-P		Use a concentration of 0.5 - 1 µg/ml.

Target

Function	Spindle assembly factor. Required for normal assembly of mitotic spindles. Required for normal assembly of microtubules during apoptosis. Required for chromatin and/or kinetochore dependent microtubule nucleation. Mediates AURKA localization to spindle microtubules. Activates AURKA by promoting its autophosphorylation at 'Thr-288' and protects this residue against dephosphorylation.
Tissue specificity	Expressed in lung carcinoma cell lines but not in normal lung tissues.
Sequence similarities	Belongs to the TPX2 family.
Developmental stage	Exclusively expressed in proliferating cells from the transition G1/S until the end of cytokinesis.
Post-translational modifications	Phosphorylated upon DNA damage, probably by ATM or ATR.
Cellular localization	Nucleus. Cytoplasm > cytoskeleton > spindle. Cytoplasm > cytoskeleton > spindle pole. During mitosis it is strictly associated with the spindle pole and with the mitotic spindle, whereas during S and G2, it is diffusely distributed throughout the nucleus. Is released from the nucleus in apoptotic cells and is detected on apoptotic microtubules.

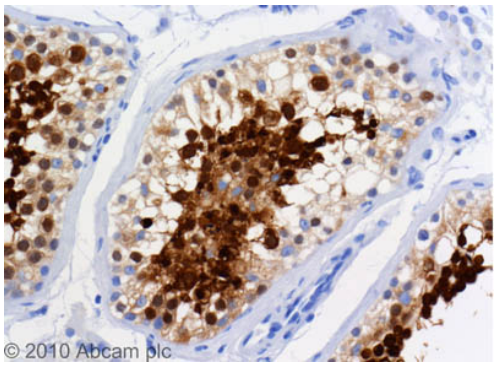
Images



Immunocytochemistry/ Immunofluorescence - TPX2 antibody [18D5-1] (ab32795)

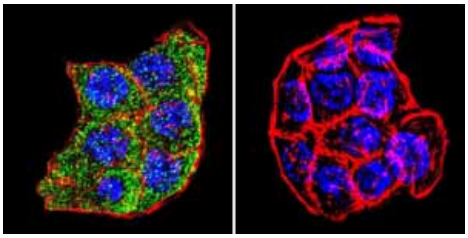
This image was kindly submitted by Serena Orlando, Giulia Guarguaglini and Patrizia Lavia, University 'La Sapienza' CNR, Italy.

ab32795 staining TPX2 in HeLa cells and mouse NIH-3T3 cells (fuzzier pattern, different from the high-quality sharp signal seen in Human cells), by immunofluorescence.
optimal antibody dilution: 4µg/ml
optimal fixation protocol: PFA/Triton fixation: 10 min room at room temperature, in 3,7 % PFA diluted in PHEM buffer (45 mM Hepes pH 6,9, 45 mM Pipes pH 6,9, 5 mM MgCl2, 10 mM EGTA) containing 0.2% Triton X-100, followed by 3 washes in PBS - Alternative fixation protocol also gives good staining: 6 min in cold Methanol at -20°C, then 3 washes in PBS.
IF was performed following a standard protocol: Blocking, 30 min; primary antibody, 1 hr; secondary antibody, 45 min. All incubations were at 37 °C in PBS/ 0.1% Tween containing 3% BSA.



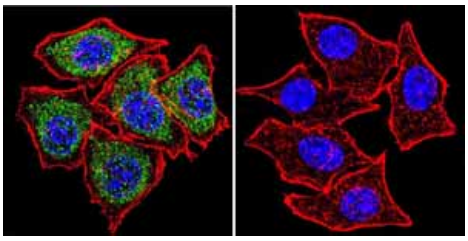
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - TPX2 antibody [18D5-1] (ab32795)

ab32795 (1µg/ml) staining TPX2 in human testis using an automated system (DAKO Autostainer Plus). Using this protocol there is strong nuclear and cytoplasmic staining . Sections were rehydrated and antigen retrieved with the Dako 3 in 1 AR buffer citrate pH6.1 in a DAKO PT link. Slides were peroxidase blocked in 3% H2O2 in methanol for 10 mins. They were then blocked with Dako Protein block for 10 minutes (containing casein 0.25% in PBS) then incubated with primary antibody for 20 min and detected with Dako envision flex amplification kit for 30 minutes. Colorimetric detection was completed with Diaminobenzidine for 5 minutes. Slides were counterstained with Haematoxylin and coverslipped under DePeX. Please note that, for manual staining, optimization of primary antibody concentration and incubation time is recommended. Signal amplification may be required.



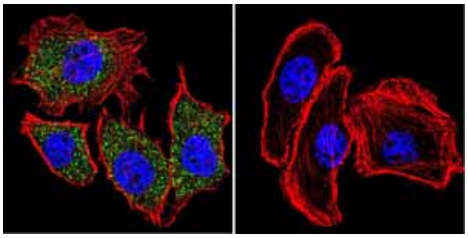
Immunocytochemistry/ Immunofluorescence-Anti-TPX2 antibody [18D5-1](ab32795)

Immunofluorescent analysis of PLK1 using PLK1 Monoclonal antibody (13E8) ab32795 shows staining in WiDr colon carcinoma cells. PLK1 staining (green) F-Actin staining with Phalloidin (red) and nuclei with DAPI (blue) is shown. Cells were grown on chamber slides and fixed with formaldehyde prior to staining. Cells were probed without (control) or with or an antibody recognizing PLK1 ab32795 at a dilution of 1:20 over night at 4 ?C washed with PBS and incubated with a DyLight-488 conjugated secondary antibody. Images were taken at 60X magnification.



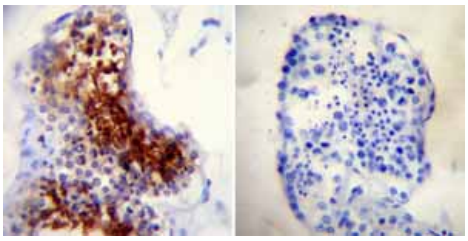
Immunocytochemistry/ Immunofluorescence-Anti-TPX2 antibody [18D5-1](ab32795)

Immunofluorescent analysis of PLK1 using PLK1 Monoclonal antibody (13E8) ab32795 shows staining in HeLa cells. PLK1 staining (green) F-Actin staining with Phalloidin (red) and nuclei with DAPI (blue) is shown. Cells were grown on chamber slides and fixed with formaldehyde prior to staining. Cells were probed without (control) or with or an antibody recognizing PLK1 ab32795 at a dilution of 1:20 over night at 4 ?C washed with PBS and incubated with a DyLight-488 conjugated secondary antibody. Images were taken at 60X magnification.



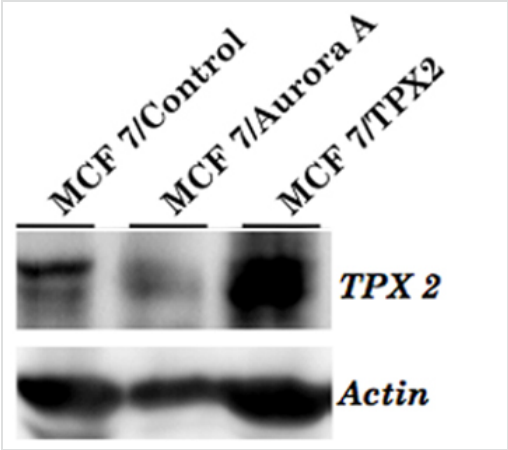
Immunocytochemistry/ Immunofluorescence-Anti-TPX2 antibody [18D5-1](ab32795)

Immunofluorescent analysis of PLK1 using PLK1 Monoclonal antibody (13E8) ab32795 shows staining in U251 glioma cells. PLK1 staining (green) F-Actin staining with Phalloidin (red) and nuclei with DAPI (blue) is shown. Cells were grown on chamber slides and fixed with formaldehyde prior to staining. Cells were probed without (control) or with or an antibody recognizing PLK1 ab32795 at a dilution of 1:20 over night at 4 ?C washed with PBS and incubated with a DyLight-488 conjugated secondary antibody. Images were taken at 60X magnification.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections)-Anti-TPX2 antibody [18D5-1] (ab32795)

Immunohistochemistry was performed on biopsies of deparaffinized Human testis tissue. To expose target proteins heat induced antigen retrieval was performed using 10mM sodium citrate (pH6.0) buffer microwaved for 8-15 minutes. Following antigen retrieval tissues were blocked in 3% BSA-PBS for 30 minutes at room temperature. Tissues were then probed at a dilution of 1:200 with a mouse monoclonal antibody recognizing TPX2 ab32795 or without primary antibody (negative control) overnight at 4°C in a humidified chamber. Tissues were washed extensively with PBST and endogenous peroxidase activity was quenched with a peroxidase suppressor. Detection was performed using a biotin-conjugated secondary antibody and SA-HRP followed by colorimetric detection using DAB. Tissues were counterstained with hematoxylin and prepped for mounting.



Western blot - Anti-TPX2 antibody [18D5-1] (ab32795)

Image from Grover A et al.,PLoS One. 2012;7(1):e30890. Epub 2012 Jan 27. Fig 7.; doi:10.1371/journal.pone.0030890; January 27, 2012, PLoS ONE 7(1): e30890.

All lanes : Anti-TPX2 antibody [18D5-1] (ab32795)

Lane 1 : MCF7 cells
Lane 2 : MCF7 cells overexpressing Aurora A
Lane 3 : MCF7 cells overexpressing TPX2

Lysates/proteins at 20 µg per lane.

Secondary
All lanes : HRP-conjugated donkey anti-mouse IgG

Developed using the ECL technique.

Predicted band size: 86 kDa

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