abcam

ab109374

Anti-Topoisomerase I antibody [EPR5375]

Anti-Topoisomerase I antibody [EPR5375] (ab109374) is a rabbit monoclonal antibody detecting Topoisomerase I in **Western Blot, Flow Cytometry (Intra), IHC-P, ICC/IF, ChIC/CUT&RUN sequencing**. Suitable for **Human, Mouse, Rat**.

- Biophysical QC for unrivalled batch-batch consistency
- Over 40 publications

Recombinant | RabMAb | Advanced Validation | 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	EPR5375
Purification technique	Affinity purification Protein A
Concentration	0.13 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IHC-P

Tested

Species	Mouse

Dilution info	1/100 - 1/250
Notes	Perform heat-mediated antigen retrieval before commencing with IHC staining protocol.

Species	Rat
Dilution info	1/100 - 1/250
Notes	Perform heat-mediated antigen retrieval before commencing with IHC staining protocol.

Species	Human
Dilution info	1/100 - 1/250
Notes	Perform heat-mediated antigen retrieval before commencing with IHC staining protocol.

IP

Not recommended

Species	Mouse, Rat, Human
Dilution info	-
Notes	-

WB

Tested

Species	Human
Dilution info	1/10000 - 1/50000
Notes	-

Expected

Species	Mouse, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	1/500
Notes	**For unpurified use at 1/100 - 1/250**

Expected

Species	Mouse, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	1/10 - 1/100
Notes	ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

Expected

Species	Mouse, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

ChIC/CUT&RUN-seq

Tested

Species	Human
Dilution info	5 µg
Notes	-

Expected

Species	Mouse, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Target data

See full target information TOP1

Function	Releases the supercoiling and torsional tension of DNA introduced during the DNA replication and transcription by transiently cleaving and rejoining one strand of the DNA duplex. Introduces a single-strand break via transesterification at a target site in duplex DNA. The scissile phosphodiester is attacked by the catalytic tyrosine of the enzyme, resulting in the formation of a DNA-(3'-phosphotyrosyl)-enzyme intermediate and the expulsion of a 5'-OH DNA strand. The free DNA strand then rotates around the intact phosphodiester bond on the opposing strand, thus removing DNA supercoils. Finally, in the religation step, the DNA 5'-OH attacks the covalent intermediate to expel the active-site tyrosine and restore the DNA phosphodiester backbone (By similarity). Regulates the alternative splicing of tissue factor (F3) pre-mRNA in endothelial cells. Involved in the circadian transcription of the core circadian clock component BMAL1 by altering the chromatin structure around the ROR response elements (ROREs) on the BMAL1 promoter.

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	Stable for 12 months at -20°C

Notes

What is this antibody validated in?

Anti-Topoisomerase I antibody [EPR5375] (ab109374) is a rabbit recombinant monoclonal antibody and is validated for use in Western Blot (WB), Flow Cytometry (Intra), Immunohistochemistry (IHC-P),

Immunocytochemistry/immunofluorescence (ICC/IF), ChIC/CUT&RUN sequencing in Human, Mouse, Rat samples.

What is the molecular weight of Topoisomerase I?
Anti-Topoisomerase I [EPR5375] (ab109374) specifically detects a band for Topoisomerase I (UniProt: P11387) at a molecular weight of 91kDa.

Trusted by the scientific community
Anti-Topoisomerase I [EPR5375] (ab109374) was first used in a scientific publication in 2011 and has been cited over 40 times in peer-reviewed journals.

Reviewed by scientists
Anti-Topoisomerase I [EPR5375] (ab109374) has over 5 independent reviews from customers.

Other related products
We have a range of other formats of antibody clone [EPR5375] also available for your convenience: ab109374, HRP - ab196642, Alexa Fluor® 647 - ab196930, Alexa Fluor® 488 - ab197505, Carrier free - ab219735

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.

Trial sizes available!
Test your antibody or perform pre-screening before committing to a larger quantity. Sold in 20µl. Discover our selection of trial-size antibodies.

Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

Activity summary	Topoisomerase I also known as DNA topoisomerase I or Topo I is an important enzyme in the unwinding of the DNA double helix during processes such as replication and transcription. This enzyme has a molecular mass of approximately 91 kDa and functions by creating transient single-strand breaks in the DNA allowing relaxation of supercoils. Topoisomerase I is widely expressed in both proliferating and non-proliferating cells with higher levels seen in tissues with rapid cell division like the gastrointestinal tract and some immune cells.
Biological function summary	Topoisomerase I plays a significant role in the modulation of DNA topology ensuring proper chromosomal functions during cellular proliferation. It serves as a single polypeptide and does not require a companion for its activity unlike other topoisomerases that may function in complexes. The enzyme's ability to relieve

	torsional stress in DNA is essential for maintaining genomic stability and facilitating the smooth progression of the replication fork.
Pathways	Topoisomerase I is vital for DNA replication and transcription processes. It works in concert with other proteins such as helicases and ligases to ensure efficient unwinding and rewinding of DNA strands. The enzyme participates in the DNA damage response pathway where it interacts with proteins like PARP1 to coordinate repair processes. Its action is important in both the S phase of the cell cycle where DNA synthesis occurs and in the G0 phase where cells are in a quiescent state.
Associated diseases and disorders	Topoisomerase I is heavily implicated in oncogenesis with significant associations to colorectal and ovarian cancers. Its heightened activity can lead to genomic instability a hallmark of cancer development. The enzyme also relates to chemotherapeutic resistance where mutations in topoisomerase I lead to reduced drug efficacy. Furthermore inhibitors targeting topoisomerase I such as camptothecin-based drugs exploit its role in cancer aiming to induce DNA damage selectively in rapidly dividing cells. Connections to other proteins like BRCA1 are apparent in these pathways as they both contribute to the DNA repair processes in cells.

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

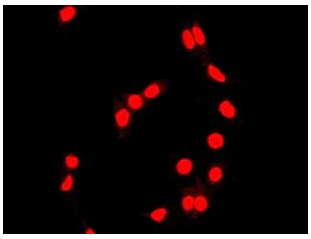
We do not recommend this combination. It is not covered by our product promise.

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.

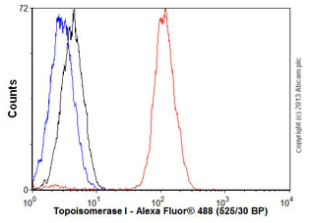
16 product images



Immunocytochemistry/ Immunofluorescence - Anti-Topoisomerase I antibody [EPR5375] (ab109374)

Topoisomerase I Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-Topoisomerase I antibody

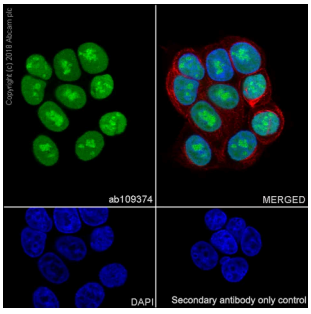
Immunofluorescent staining of MCF7 cells using ab109374 at a dilution of 1/100.



Flow Cytometry (Intracellular) - Anti-Topoisomerase I antibody [EPR5375] (ab109374)

Topoisomerase I Flow Cytometry (Intracellular) staining using rabbit Anti-Topoisomerase I antibody

Overlay histogram showing HepG2 cells stained with ab109374 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween 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 (ab109374, 1/100 dilution) for 30 min at 22°C. The secondary antibody used was Alexa Fluorr® 488 goat anti-rabbit IgG (H&L) (ab150077) at 1/2000 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. Unlabelled sample (blue line) was also used as a control. Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter.



Immunocytochemistry/ Immunofluorescence - Anti-Topoisomerase I antibody [EPR5375] (ab109374)

Topoisomerase I Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-Topoisomerase I antibody

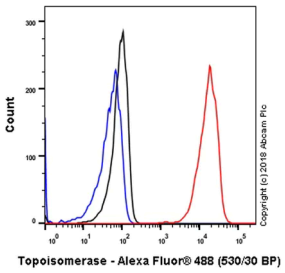
Immunocytochemistry/ Immunofluorescence analysis of MCF7 (Human breast adenocarcinoma epithelial cell) cells labeling Topoisomerase I with Purified ab109374 at 1/500 dilution (0.3 µg/ml). Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% tritonX-100. Cells were counterstained with ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1:200 (2.5 µg/ml). Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) was used as the secondary antibody at 1/1000 (2 µg/ml) dilution. DAPI nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Topoisomerase I antibody [EPR5375] (ab109374)

Topoisomerase I Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-Topoisomerase I antibody

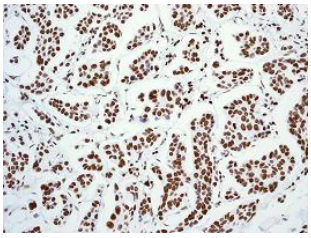
Immunohistochemical analysis of paraffin-embedded Human colonic carcinoma tissue using ab109374 at a dilution of 1/100. Heat mediated antigen retrieval was performed before commencing with IHC staining protocol.



Flow Cytometry (Intracellular) - Anti-Topoisomerase I antibody [EPR5375] (ab109374)

Topoisomerase I Flow Cytometry (Intracellular) staining using rabbit Anti-Topoisomerase I antibody

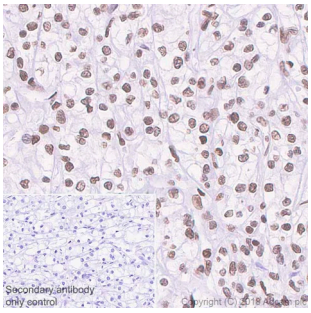
Intracellular Flow Cytometry analysis of HepG2 (Human hepatocellular carcinoma epithelial cell) cells labeling Topoisomerase I with Purified ab109374 at 1/20 dilution (10µg/ml) (red). Cells were fixed with 80% Methanol. A Goat anti rabbit IgG (Alexa Fluorr® 488, ab150077) secondary antibody was used at 1/2000. Isotype control - Rabbit monoclonal IgG (Black). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Topoisomerase I antibody [EPR5375] (ab109374)

Topoisomerase I Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-Topoisomerase I antibody

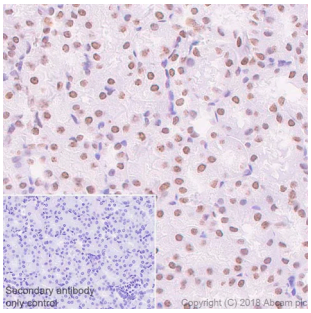
Immunohistochemical analysis of paraffin-embedded Human breast carcinoma tissue using ab109374 at a dilution of 1/100. Heat mediated antigen retrieval was performed before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Topoisomerase I antibody [EPR5375] (ab109374)

Topoisomerase I Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-Topoisomerase I antibody

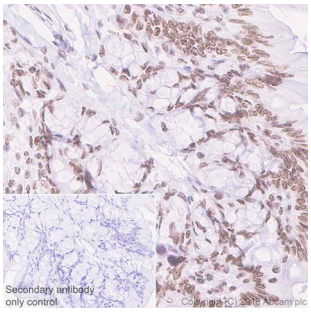
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human clear cell carcinoma of kidney tissue sections labeling Topoisomerase I with Purified ab109374 at 1:100 dilution (1.29 µg/ml). Heat mediated antigen retrieval was performed using ab93684 (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use)was used as the secondary antibody. Negative control:PBS instead of the primary antibody. Hematoxylin was used as a counterstain.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Topoisomerase I antibody [EPR5375] (ab109374)

Topoisomerase I Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-Topoisomerase I antibody

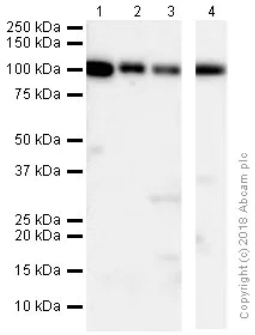
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Mouse kidney tissue sections labeling Topoisomerase I with purified ab109374 at 1:100 dilution (1.29 µg/ml). Heat mediated antigen retrieval was performed using ab93684 (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use)was used as the secondary antibody. Negative control:PBS instead of the primary antibody. Hematoxylin was used as a counterstain.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Topoisomerase I antibody [EPR5375] (ab109374)

Topoisomerase I Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-Topoisomerase I antibody

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Rat colon tissue sections labeling Topoisomerase I with purified ab109374 at 1:100 dilution (1.29 µg/ml). Heat mediated antigen retrieval was performed using ab93684 (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use)was used as the secondary antibody. Negative control:PBS instead of the primary antibody. Hematoxylin was used as a counterstain.



Western blot - Anti-Topoisomerase I antibody [EPR5375] (ab109374)

All lanes:
Western blot - Anti-Topoisomerase I antibody [EPR5375] (ab109374) at 1/10000 dilution

Lane 1:
MCF7 (Human breast adenocarcinoma epithelial cell) whole cell lysates at 20 µg

Lane 2:
Jurkat (Human T cell leukemia T lymphocyte) whole cell lysates at 20 µg

Lane 3:
HepG2 (Human hepatocellular carcinoma epithelial cell) whole cell lysates at 20 µg

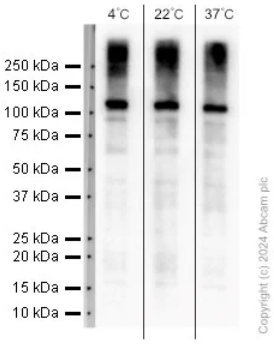
Lane 4:
Neuro-2a (Mouse neuroblastoma neuroblast) whole cell lysates at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

Predicted band size: 91 kDa

Observed band size: 91 kDa



Western blot - Anti-Topoisomerase I antibody [EPR5375] (ab109374)

Topoisomerase I Western blot staining of Jurkat whole cell lysate using rabbit Anti-Topoisomerase I antibody

This blot was produced using 4-20% SDS-PAGE containing 15 µg of Jurkat whole cell lysate per lane at 150V for 1hr before being transferred onto a 0.45 µm PVDF membrane at 75V for 1hr. The membrane was then blocked for 1hr using 5% NFDM/TBST, then incubated with ab109374 (1/10,000) at room temperature for 1hr. After being washed three times in TBST, the membrane was incubated with Peroxidase conjugated goat anti-rabbit IgG (H+L) (ab97051) at 1/20,000 dilution for 1hr at room temperature. The membrane was washed three times again. Then the signal was developed using the ECL technique.

ab109374 was stored at a range of temperatures (+4°C, +22°C, +37°C) for 1 week before being tested in WB. The image shows the band intensity remains relatively constant across all storage temperatures, demonstrating that antibody activity is not affected under these conditions. This data was generated as part of a structured antibody stability study supporting Abcam’ s transition from cold⊠chain to ambient shipping.

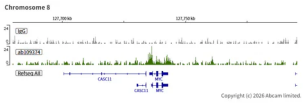
All lanes:
Western blot - Anti-Topoisomerase I antibody [EPR5375] (ab109374) at 1/10000 dilution

All lanes:
Jurkat whole cell lysate at 15 µg with NDFM/TBST

Secondary

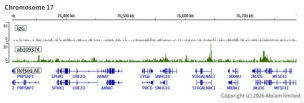
All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

Exposure time: 60s



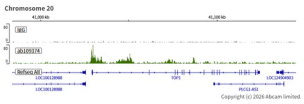
ChIC/CUT&RUN sequencing - Anti-Topoisomerase I antibody [EPR5375] (ab109374)

ChIC/CUT&RUN was performed using a pAG-MNase at a final concentration of 700 ng/mL, 2.5 x 10^5 K-562 (human chronic myelogenous leukemia lymphoblast) cells and 5 µg of ab109374 [EPR5375]. The resulting DNA was sequenced on the Illumina NovaSeq 6000 to a depth of 10 million reads. The negative IgG control ab172730 is also shown. The University of Geneva owns patents relevant to ChIC (Chromatin Immuno-Cleavage) methods.



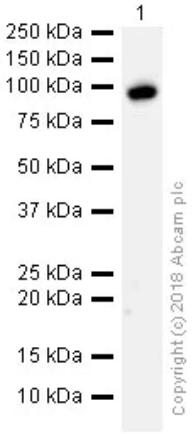
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Western blot - Anti-Topoisomerase I antibody [EPR5375] (ab109374)

Blocking and diluting buffer and concentration: 5% NFDM/TBST
All lanes:
Western blot - Anti-Topoisomerase I antibody [EPR5375] (ab109374) at 1/50000 dilution

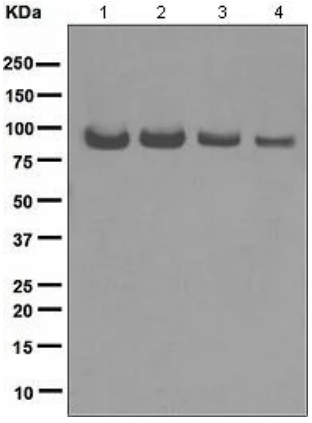
All lanes:
PC-12 (Rat adrenal gland pheochromocytoma) whole cell lysates at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

Predicted band size: 91 kDa

Observed band size: 91 kDa



Western blot - Anti-Topoisomerase I antibody [EPR5375] (ab109374)

All lanes:
Western blot - Anti-Topoisomerase I antibody [EPR5375] (ab109374) at 1/10000 dilution

Lane 1:
Jurkat cell lysate at 10 µg

Lane 2:
MCF7 cell lysate at 10 µg

Lane 3:
SW480 cell lysate at 10 µg

Lane 4:
K562 cell lysate at 10 µg

Predicted band size: 91 kDa

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