

Anti-Topoisomerase II alpha/TOP2A Antibody Picoband®

Catalog Number: A00953

About TOP2A

The human topoisomerase II enzyme is encoded by a single-copy gene which is mapped to 17q21-q22. The TOP2A gene spans approximately 30 kb and contains 35 exons. Furthermore, DNA topoisomerases are enzymes that control and alter the topologic states of DNA in both prokaryotes and eukaryotes. Topoisomerase II from eukaryotic cells catalyzes the relaxation of supercoiled DNA molecules, catenation, decatenation, knotting, and unknotting of circular DNA. It appears likely that the reaction catalyzed by topoisomerase II involves the crossing-over of 2 DNA segments. There are about 100,000 molecules of topoisomerase II per HeLa cell nucleus, constituting about 0.1% of the nuclear extract¹. DNA topoisomerase II-alpha is associated with the pol II holoenzyme and is a required component of chromatin-dependent coactivation. Specific inhibitors of topoisomerase II blocked transcription on chromatin templates, but did not affect transcription on naked templates. Addition of purified topoisomerase II-alpha reconstituted chromatin-dependent activation activity in reactions with core pol II².

Overview

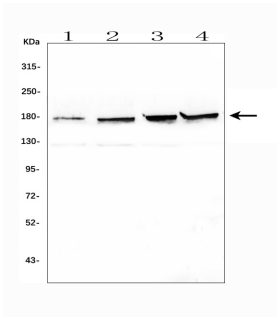
Product Name	Anti-Topoisomerase II alpha/TOP2A Antibody Picoband®
Reactive Species	Human
Description	Boster Bio Anti-Topoisomerase II alpha/TOP2A Antibody Picoband® catalog # A00953. Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human. The brand Picoband indicates this is a premium antibody that guarantees superior quality, high affinity, and strong signals with minimal background in Western blot applications. Only our best-performing antibodies are designated as Picoband, ensuring unmatched performance.
Application	Flow Cytometry, IF, IHC, ICC, WB
Clonality	Polyclonal
Formulation	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na ₂ HPO ₄ , 0.05mg NaN ₃ .
Storage Instructions	Store at -20°C for one year from date of receipt. After reconstitution, at 4°C for one month. It can also be aliquotted and stored frozen at -20°C for six months. Avoid repeated freeze-thaw cycles.
Host	Rabbit
Uniprot ID	P11388

Technical Details

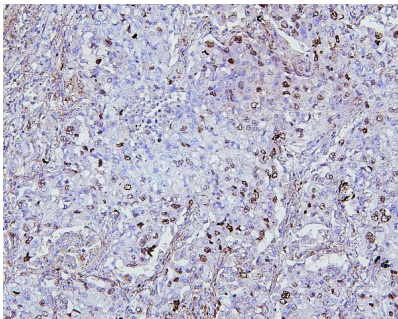
Immunogen	A synthetic peptide corresponding to a sequence in the middle region of human Topoisomerase II alpha/TOP2A, identical to the related mouse and rat sequences.
Recommended Detection Systems	Boster recommends Enhanced Chemiluminescent Kit with anti-Rabbit IgG (EK1002) for Western blot, and HRP Conjugated anti-Rabbit IgG Super Vision Assay Kit (SV0002-1) for IHC(P) and ICC.
Cross Reactivity	No cross-reactivity with other proteins.

Isotype	Rabbit IgG
Form	Lyophilized
Concentration	Adding 0.2 ml of distilled water will yield a concentration of 500 ug/ml.
Purification	Immunogen affinity purified.
Suggested Dilutions	Western blot, 0.25-0.5ug/ml, Human Immunohistochemistry (Paraffin-embedded Section), 0.5-1ug/ml, Human Immunocytochemistry/Immunofluorescence, 2ug/ml, Human Flow Cytometry (Fixed), 1-3ug/1x10 ⁶ cells, Human

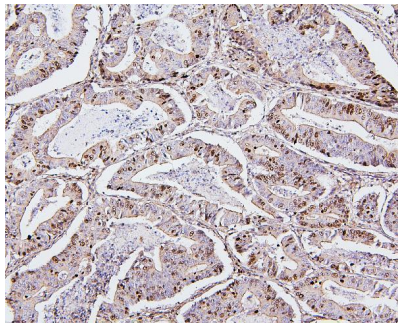
Anti-Topoisomerase II alpha/TOP2A Antibody Picoband® (A00953) Images



Western blot analysis of TOP2A using anti-TOP2A antibody (A00953). Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions. Lane 1: human A549 whole cell lysates, Lane 2: human U2OS whole cell lysates, Lane 3: human HEK293 whole cell lysates, Lane 4: human K562 whole cell lysates. After Electrophoresis, proteins were transferred to a Nitrocellulose membrane at 150mA for 50-90 minutes. Blocked the membrane with 5% Non-fat Milk/TBS for 1.5 hour at RT. The membrane was incubated with rabbit anti-TOP2A antigen affinity purified polyclonal antibody (Catalog # A00953) at 0.5 ug/mL overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-rabbit IgG-HRP secondary antibody at a dilution of 1:10000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1002) with Tanon 5200 system. A specific band was detected for TOP2A at approximately 174KD. The expected band size for TOP2A is at 174KD.

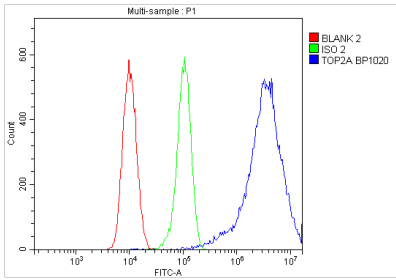


IHC analysis of TOP2A using anti-TOP2A antibody (A00953). TOP2A was detected in paraffin-embedded section of human Lung cancer tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml rabbit anti-TOP2A Antibody (A00953) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC)(Catalog # SA1022) with DAB as the chromogen.

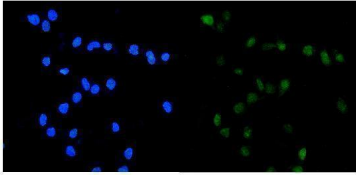


IHC analysis of TOP2A using anti-TOP2A antibody (A00953). TOP2A was detected in paraffin-embedded section of human rectal cancer tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml rabbit anti-TOP2A Antibody (A00953) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC)(Catalog # SA1022) with DAB as the chromogen.

Flow Cytometry analysis of 293T cells using anti-TOP2A antibody (A00953). Overlay histogram showing 293T cells stained with A00953 (Blue line). To facilitate intracellular staining, cells were fixed with 4% paraformaldehyde and permeabilized with permeabilization buffer. The cells were blocked with 10% normal goat serum. And then incubated



with rabbit anti-TOP2A Antibody (A00953, 1 μ g/1x10⁶ cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (BA1127, 5-10 μ g/1x10⁶ cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1 μ g/1x10⁶) used under the same conditions. Unlabelled sample without incubation with primary antibody and secondary antibody (Red line) was used as a blank control.



IF analysis of TOP2A using anti-TOP2A antibody (A00953). TOP2A was detected in immunocytochemical section of U2OS cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 2 μ g/mL rabbit anti-TOP2A Antibody (A00953) overnight at 4°C. DyLight®488 Conjugated Goat Anti-Rabbit IgG (BA1127) was used as secondary antibody at 1:100 dilution and incubated for 30 minutes at 37°C. The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.

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