abcam

Product datasheet

Anti-Topoisomerase II beta/TOP2B antibody ab125297

KO VALIDATED

2 References 3 Images

Overview

Product name Anti-Topoisomerase II beta/TOP2B antibody

Description Rabbit polyclonal to Topoisomerase II beta/TOP2B

Host species Rabbit

Tested applications Suitable for: WB

Species reactivity Reacts with: Mouse, Human

Predicted to work with: Rat, Cow, Dog, Pig, Chimpanzee, Macaque monkey, Gorilla,

Orangutan 4

Immunogen Synthetic peptide corresponding to Human Topoisomerase II beta/TOP2B aa 1-100 conjugated

to keyhole limpet haemocyanin. (Peptide available as <u>ab156303</u>)

Positive control WB: Wild-type NALM-6 cell lysate. HeLa and NIH/3T3 whole cell lysate. Jurkat, K562, MCF7 and

HepG2 cell lysate.

General notesThe 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 short term (1-2 weeks). Upon delivery aliquot. Store at -20°C or -

80°C. Avoid freeze / thaw cycle.

Storage buffer pH: 7.40

Preservative: 0.02% Sodium azide

Constituent: PBS

Batches of this product that have a concentration < 1mg/ml may have BSA added as a stabilising

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agent. If you would like information about the formulation of a specific lot, please contact our

scientific support team who will be happy to help.

Purity Immunogen affinity purified

Clonality Polyclonal

Isotype IgG

Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab125297 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
WB		Use a concentration of 1 µg/ml. Detects a band of approximately 183 kDa (predicted molecular weight: 183 kDa). Abcam recommends using milk as the blocking agent - 5%

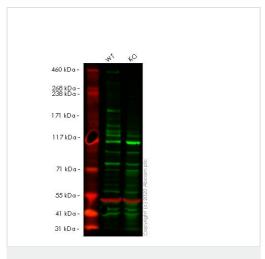
Function	Control of topological states of DNA by transient breakage and subsequent rejoining of DNA	
	strands. Topoisomerase II makes double-strand breaks. Indirectly ivolved in vitamin D-coupled	
	transcription regulation via its association with the WINAC complex, a chromatin-remodeling	
	complex recruited by vitamin D receptor (VDR), which is required for the ligand-bound VDR-	
	mediated transrepression of the CYP27B1 gene.	
Sequence similarities	Belongs to the type II topoisomerase family.	
Post-translational modifications	Phosphorylated upon DNA damage, probably by ATM or ATR.	

Cytoplasm. Nucleus > nucleolus.

Images

Cellular localization

Target



Western blot - Anti-Topoisomerase II beta/TOP2B antibody (ab125297)

All lanes : Anti-Topoisomerase II beta/TOP2B antibody (ab125297)

Lane 1: Wild-type HEK-293T cell lysate

Lane 2: TOP2B knockout HEK-293T cell lysate

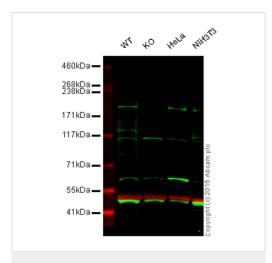
Lysates/proteins at 40 µg per lane.

Performed under reducing conditions.

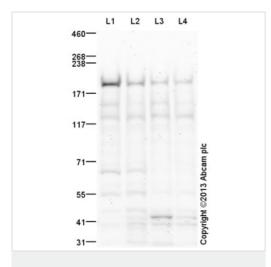
Predicted band size: 183 kDa Observed band size: 183 kDa

Lanes 1-2: Merged signal (red and green). Green - ab125297 observed at 183 kDa. Red - Anti-alpha Tubulin antibody [DM1A] - Loading Control (ab7291) observed at 50 kDa.

ab125297 was shown to react with Topoisomerase II beta/TOP2B in wild-type HEK-293T cells in western blot. Loss of signal was observed when knockout cell line ab266340 (knockout cell lysate ab257286) was used. Wild-type HEK-293T and TOP2B knockout HEK-293T cell lysates were subjected to SDS-PAGE. ab125297 and Anti-alpha Tubulin antibody [DM1A] - Loading Control (ab7291) overnight at 4°C at a 1 µg/ml and a 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye®800CW) preadsorbed (ab216773) and Goat anti-Mouse lgG H&L (IRDye®680RD) preadsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-Topoisomerase II beta/TOP2B antibody (ab125297)



Western blot - Anti-Topoisomerase II beta/TOP2B antibody (ab125297)

Lane 1: Wild-type NALM-6 cell lysate (20 µg)

Lane 2: Topoisomerase II beta/TOP2B knockout NALM-6 cell lysate (20 μg)

Lane 3: HeLa cell lysate (20 µg)

Lane 4: NIH/3T3 cell lysate (20 µg)

Lanes 1 - 4: Merged signal (red and green). Green - ab125297 observed at 200 kDa. Red - loading control, <u>ab7291</u>, observed at 50 kDa.

ab125297 was shown to recognize Topoisomerase II beta/TOP2B when Topoisomerase II beta/TOP2B knockout samples were used, along with additional cross-reactive bands. Wild-type and Topoisomerase II beta/TOP2B knockout samples were subjected to SDS-PAGE. ab125297 and ab7291 (loading control to alpha tubulin) were diluted 1 µg/mL and 1/2000 and incubated overnight at 4°C. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed ab216773 and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed ab216776 secondary antibodies at 1/10000 dilution for 1 h at room temperature before imaging.

All lanes : Anti-Topoisomerase II beta/TOP2B antibody (ab125297) at 1 μg/ml (Milk blocking - 5%)

Lane 1 : Jurkat (Human T cell lymphoblast-like cell line) Whole Cell Lysate

Lane 2 : K562 (Human erythromyeloblastoid leukemia cell line) Whole Cell Lysate

Lane 3: MCF7 (Human breast adenocarcinoma cell line) Whole Cell Lysate

Lane 4 : HepG2 (Human hepatocellular liver carcinoma cell line) Whole Cell Lysate

Lysates/proteins at 25 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/10000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 183 kDa **Observed band size:** 183 kDa

Additional bands at: 120 kDa, 150 kDa, 42 kDa, 55 kDa, 66 kDa.

We are unsure as to the identity of these extra bands.

Exposure time: 8 minutes

Abcam recommends using milk as the blocking agent - 5%. This blot was produced using a 3-8% Tris Acetate gel under the TA buffer system. The gel was run at 150V for 60 minutes before being transferred onto a Nitrocellulose membrane at 30V for 70 minutes. The membrane was then blocked for an hour using 5% Milk before being incubated with ab125297 overnight at 4°C. Antibody binding was detected using an anti-rabbit antibody conjugated to HRP, and

visualised using ECL development solution.

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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