abcam

Product datasheet

Anti-SA2 antibody [EPR17865] - C-terminal ab201451





★★★★★ 2 Abreviews 1 References 13 Images

Overview

Product name Anti-SA2 antibody [EPR17865] - C-terminal

Description Rabbit monoclonal [EPR17865] to SA2 - C-terminal

Host species Rabbit

Tested applications Suitable for: Flow Cyt (Intra), WB, IHC-P, ICC/IF

Species reactivity Reacts with: Mouse, Rat, Human

Immunogen Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: HeLa, HCT116, MCF-7, K562, C6, Raw264.7 and NIH3T3 cell lysates; Human fetal brain

and Mouse spleen lysates; IHC-P: Human breast carcinoma, human tonsil, mouse and rat spleen

tissue; IF: MCF-7 and K562 cells.

General notes 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 production

For more information see here.

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**® **patents**.

Properties

Liquid **Form**

Storage instructions Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long

term. Avoid freeze / thaw cycle.

Storage buffer pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

Purity Protein A purified

Clonality Monoclonal Clone number EPR17865

Isotype IgG

Applications

The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab201451 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)		1/25000.
WB	**** <u>(2)</u>	1/1000. Detects a band of approximately 141 kDa (predicted molecular weight: 141 kDa).
IHC-P		1/2000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
ICC/IF		1/500.

Target

Function

Component of cohesin complex, a complex required for the cohesion of sister chromatids after DNA replication. The cohesin complex apparently forms a large proteinaceous ring within which sister chromatids can be trapped. At anaphase, the complex is cleaved and dissociates from chromatin, allowing sister chromatids to segregate. The cohesin complex may also play a role in spindle pole assembly during mitosis.

Sequence similarities

Belongs to the SCC3 family.

Contains 1 SCD (stromalin conservative) domain.

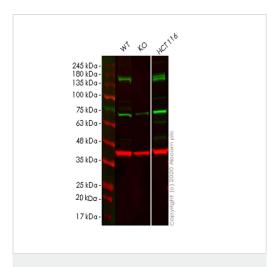
Post-translational modifications

Phosphorylated by PLK. The large dissociation of cohesin from chromosome arms during prophase is partly due to its phosphorylation.

Cellular localization

Nucleus. Chromosome. Chromosome > centromere. Associates with chromatin. Before prophase it is scattered along chromosome arms. During prophase, most of cohesin complexes dissociate from chromatin probably because of phosphorylation by PLK, except at centromeres, where cohesin complexes remain. At anaphase, the RAD21 subunit of cohesin is cleaved, leading to the dissociation of the complex from chromosomes, allowing chromosome separation. In germ cells, cohesin complex dissociates from chromatin at prophase I, and may be replaced by a meiosis-specific cohesin complex.

Images



Western blot - Anti-SA2 antibody [EPR17865] - C-terminal (ab201451)

All lanes : Anti-SA2 antibody [EPR17865] - C-terminal (ab201451) at 1/1000 dilution

Lane 1: Wild-type HeLa cell lysate

Lane 2: STAG2 knockout HeLa cell lysate

Lane 3: HCT116 cell lysate

Lysates/proteins at 20 µg per lane.

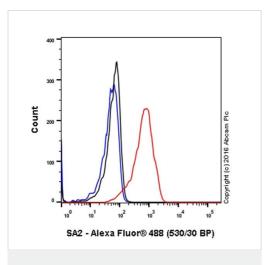
Secondary

All lanes : Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (**ab216773**) at 1/10000 dilution

Predicted band size: 141 kDa Observed band size: 141 kDa

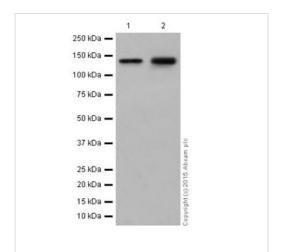
Lanes 1-3: Merged signal (red and green). Green - ab201451 observed at 141 kDa. Red - loading control <u>ab8245</u> observed at 36 kDa.

ab201451 Anti-SA2 antibody [EPR17865] - C-terminal was shown to specifically react with SA2 in wild-type HeLa cells. Loss of signal was observed when knockout cell line ab265461 (knockout cell lysate ab257707) was used. Wild-type and SA2 knockout samples were subjected to SDS-PAGE. ab201451 and Anti-GAPDH antibody [6C5] - Loading Control (ab8201451 and Anti-GAPDH antibody [6C5] - Loading Control (ab8245) were incubated overnight at 4°C at 1 in 1000 dilution and 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.



Flow Cytometry (Intracellular) - Anti-SA2 antibody [EPR17865] - C-terminal (ab201451)

Intracellular Flow Cytometry analysis of K562 (human chronic myelogenous leukemia) labelling SA2 with purified ab201451 at 1/25000 (red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. Alexa Fluor[®] 488 goat antirabbit lgG (1/2000) was used as the secondary antibody. Black-lsotype control, rabbit monoclonal lgG. Blue - Unlabelled control, cells without incubation with primary and secondary antibodies.



Western blot - Anti-SA2 antibody [EPR17865] - C-terminal (ab201451)

All lanes : Anti-SA2 antibody [EPR17865] - C-terminal (ab201451) at 1/10000 dilution

Lane 1 : MCF-7 (Human breast adenocarcinoma cell line) cell lysate

Lane 2: K562 (Human chronic myelogenous leukemia cells from bone marrow) cell lysate

Lysates/proteins at 10 µg per lane.

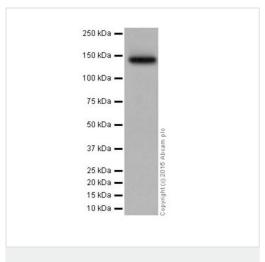
Secondary

All lanes : Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated at 1/1000 dilution

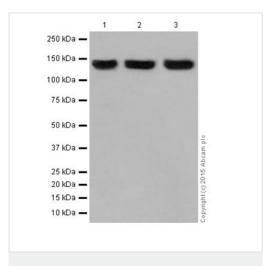
Predicted band size: 141 kDa **Observed band size:** 141 kDa

Exposure time: 3 minutes

5% NFDM/TBST: Blocking and diluting buffer.



Western blot - Anti-SA2 antibody [EPR17865] - Cterminal (ab201451)



Western blot - Anti-SA2 antibody [EPR17865] - C-terminal (ab201451)

Anti-SA2 antibody [EPR17865] - C-terminal (ab201451) at 1/1000 dilution + Human fetal brain lysate at 10 µg

Secondary

Anti-Rabbit lgG (HRP), specific to the non-reduced form of lgG at 1/1000 dilution

Predicted band size: 141 kDa **Observed band size:** 141 kDa

Exposure time: 1 minute

5% NFDM/TBST: Blocking and diluting buffer.

All lanes : Anti-SA2 antibody [EPR17865] - C-terminal (ab201451) at 1/10000 dilution

Lane 1: C6 (Rat glial tumor cells) cell lysate

Lane 2: Raw264.7 (Mouse macrophage cells transformed with

Abelson murine leukemia virus) cell lysate

Lane 3: NIH 3T3 (Mouse embyro fibroblast cells) cell lysate

Lysates/proteins at 10 µg per lane.

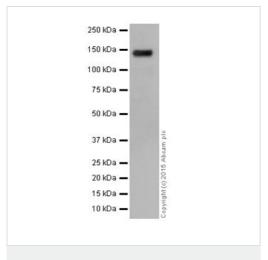
Secondary

All lanes : Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated at 1/1000 dilution

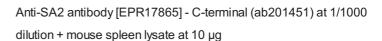
Predicted band size: 141 kDa **Observed band size:** 141 kDa

Exposure time: 3 minutes

5% NFDM/TBST: Blocking and diluting buffer.



Western blot - Anti-SA2 antibody [EPR17865] - C-terminal (ab201451)



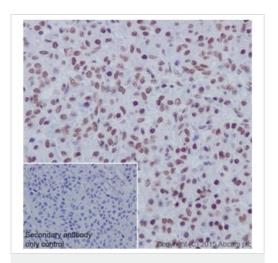
Secondary

Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/1000 dilution

Predicted band size: 141 kDa **Observed band size:** 141 kDa

Exposure time: 1 minute

5% NFDM/TBST: Blocking and diluting buffer.



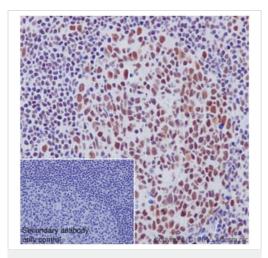
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-SA2 antibody [EPR17865] - C-terminal (ab201451)

Immunohistochemical analysis of paraffin-embedded Human breast carcinoma tissue labeling SA2 using ab201451 at 1/2000 dilution. A Goat Anti-Rabbit IgG H&L (HRP) (ab97051) was used as secondary at 1/500 dilution. Counterstain: Hematoxylin. Inset image: negative control obtained using PBS instead of ab201451, and secondary antibody only.

Note: Nuclear staining on Human breast carcinoma tissue was

observed.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-SA2 antibody

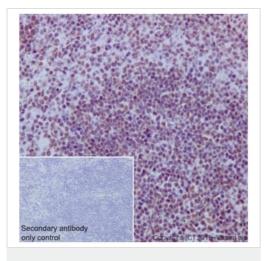
[EPR17865] - C-terminal (ab201451)

Immunohistochemical analysis of paraffin-embedded Human tonsil tissue labeling SA2 using ab201451 at 1/2000 dilution. A Goat Anti-Rabbit lgG H&L (HRP) (ab97051) was used as secondary at 1/500 dilution. Counterstain: Hematoxylin.

Inset image: negative control obtained using PBS instead of ab201451, and secondary antibody.

Note: Nuclear staining on Human tonsil tissue was observed.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-SA2 antibody

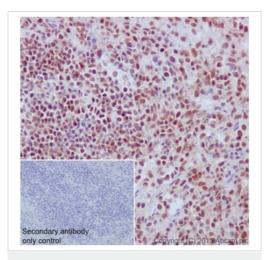
[EPR17865] - C-terminal (ab201451)

Immunohistochemical analysis of paraffin-embedded mouse spleen tissue labeling SA2 using ab201451 at 1/2000 dilution. A Goat Anti-Rabbit IgG H&L (HRP) (ab97051) was used as secondary at 1/500 dilution. Counterstain: Hematoxylin.

Inset image: negative control obtained using PBS instead of ab201451, and secondary antibody.

Note: Nuclear staining on mouse spleen tissue was observed.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-SA2 antibody

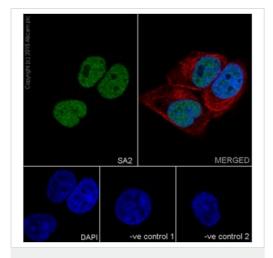
[EPR17865] - C-terminal (ab201451)

Immunohistochemical analysis of paraffin-embedded rat spleen tissue labeling SA2 using ab201451 at 1/2000 dilution. A Goat Anti-Rabbit lgG H&L (HRP) (ab97051) was used as secondary at 1/500 dilution. Counterstain: Hematoxylin.

Inset image: negative control obtained using PBS instead of ab201451, and secondary antibody.

Note: Nuclear staining on rat spleen tissue was observed.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunocytochemistry/ Immunofluorescence - Anti-SA2 antibody [EPR17865] - C-terminal (ab201451)

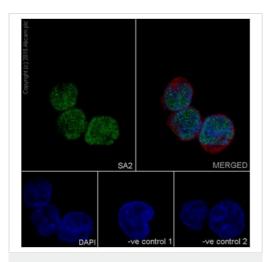
Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized MCF7 (Human breast adenocarcinoma cell line) cells labeling SA2 with ab201451 at 1/500 dilution, followed by Goat anti-rabbit lgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/500 dilution (green).

Confocal image showing nuclear staining on MCF7 cell line.

The nuclear counter stain is DAPI (blue). Tubulin is detected with ab7291 (anti-Tubulin mouse mAb) at 1/1000 dilution and ab150120 (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution (red).

The negative controls are as follows:

ab201451 at 1/500 dilution followed by <u>ab150120</u>
 (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution.
 <u>ab7291</u> (anti-Tubulin mouse mAb) at 1/1000 dilution followed by <u>ab150077</u> (Alexa Fluor®488 Goat Anti-Rabbit lgG H&L) at 1/500 dilution.



Immunocytochemistry/ Immunofluorescence - Anti-SA2 antibody [EPR17865] - C-terminal (ab201451)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized K562 (Human chronic myelogenous leukemia cells from bone marrow) cells labeling SA2 with ab201451 at 1/500 dilution, followed by Goat anti-rabbit lgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/500 dilution (green).

Confocal image showing nuclear staining on K562 cell line.

The nuclear counter stain is DAPI (blue). Tubulin is detected with <u>ab7291</u> (anti-Tubulin mouse mAb) at 1/1000 dilution and <u>ab150120</u> (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution (red).

The negative controls are as follows:

1. ab201451 at 1/500 dilution followed by <u>ab150120</u> (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution.
2. <u>ab7291</u> (anti-Tubulin mouse mAb) at 1/1000 dilution followed by <u>ab150077</u> (Alexa Fluor®488 Goat Anti-Rabbit lgG H&L) at 1/500 dilution.



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