abcam

Product datasheet

Anti-Rad50 antibody [EPR20968] - ChIP Grade ab208019

Recombinant RabMAb

9 Images

Overview

Product name	Anti-Rad50 antibody [EPR20968] - ChIP Grade
Description	Rabbit monoclonal [EPR20968] to Rad50 - ChIP Grade
Host species	Rabbit
Tested applications	**Suitable for:** WB, ChIP, IHC-P
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: Human fetal brain and kidney lysates; K562, HeLa, HEK-293T, HT-29, NIH/3T3, ES-D3, C6, RAW 264.7 and PC-12 cell lysates; Mouse kidney lysate; Rat brain and spleen lysate. IHC-P: Human breast and breast carcinoma tissue; Mouse and rat testis tissue. ChIP: Chromatin prepared from HeLa 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

Form	Liquid
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: 7.2 Preservative: 0.01% Sodium azide Constituents: PBS, 0.05% BSA, 40% Glycerol (glycerin, glycerine)
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR20968

Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab208019 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		1/1000. Detects a band of approximately 154 kDa (predicted molecular weight: 153 kDa).
ChIP		Use 2 µg for 25 µg of chromatin.
IHC-P		1/4000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Target

Function	Component of the MRN complex, which plays a central role in double-strand break (DSB) repair, DNA recombination, maintenance of telomere integrity and meiosis. The complex possesses single-strand endonuclease activity and double-strand-specific 3'-5' exonuclease activity, which are provided by MRE11A. RAD50 may be required to bind DNA ends and hold them in close proximity. This could facilitate searches for short or long regions of sequence homology in the recombining DNA templates, and may also stimulate the activity of DNA ligases and/or restrict the nuclease activity of MRE11A to prevent nucleolytic degradation past a given point. The complex may also be required for DNA damage signaling via activation of the ATM kinase. In telomeres the MRN complex may modulate t-loop formation.
Tissue specificity	Expressed at very low level in most tissues, except in testis where it is expressed at higher level. Expressed in fibroblasts.
Involvement in disease	Defects in RAD50 are the cause of Nijmegen breakage syndrome-like disorder (NBSLD) [MIM:613078]; also called NBS-like disorder or RAD50 deficiency. NBSLD is a disorder similar to Nijmegen breakage syndrome and characterized by chromosomal instability, radiation sensitivity, microcephaly, growth retardation, short stature and bird-like face. Immunodeficiency is absent.
Sequence similarities	Belongs to the SMC family. RAD50 subfamily. Contains 1 zinc-hook domain.
Domain	The zinc-hook, which separates the large intramolecular coiled coil regions, contains 2 Cys residues that coordinate one molecule of zinc with the help of the 2 Cys residues of the zinc-hook of another RAD50 molecule, thereby forming a V-shaped homodimer. The two heads of the homodimer, which constitute the ATP-binding domain, interact with the MRE11A homodimer.
Post-translational modifications	Phosphorylated upon DNA damage, probably by ATM or ATR.
Cellular localization	Nucleus. Chromosome > telomere. Localizes to discrete nuclear foci after treatment with genotoxic agents.

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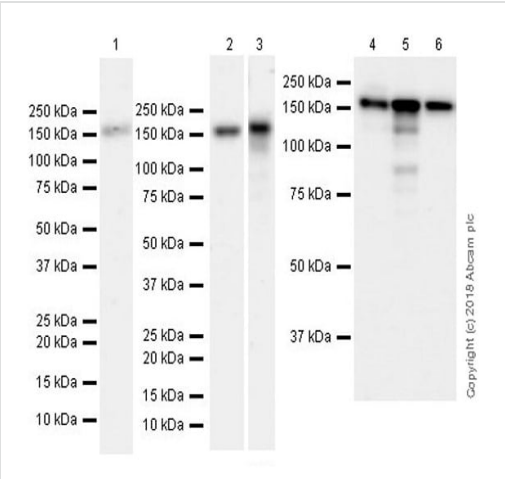
Images



Western blot - Anti-Rad50 antibody [EPR20968] - ChIP Grade (ab208019)

All lanes : Anti-Rad50 antibody [EPR20968] - ChIP Grade (ab208019) at 1/1000 dilution

Lane 1 : Mouse kidney lysate at 10 µg
Lane 2 : Rat brain lysate at 10 µg
Lane 3 : Rat spleen lysate at 10 µg
Lane 4 : C6 (rat glial tumor cell line) whole cell lysate at 20 µg
Lane 5 : RAW 364.7 (mouse macrophage cell line transformed with Abelson murine leukemia virus) whole cell lysate at 20 µg
Lane 6 : PC-12 (rat adrenal gland pheochromocytoma cell line) whole cell lysate at 20 µg

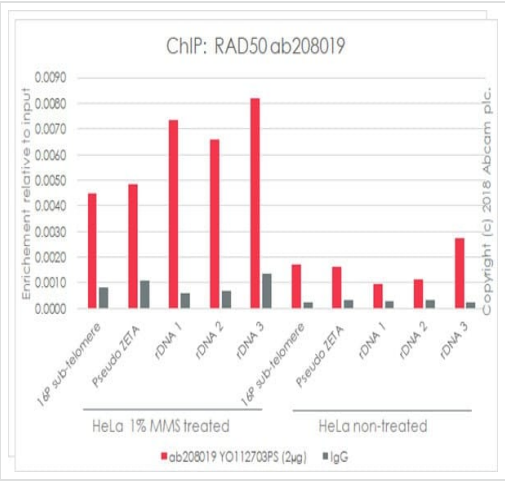
Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/100000 dilution

Predicted band size: 153 kDa
Observed band size: 154 kDa

Blocking/Diluting buffer and concentration: 5% NFDM/TBST.

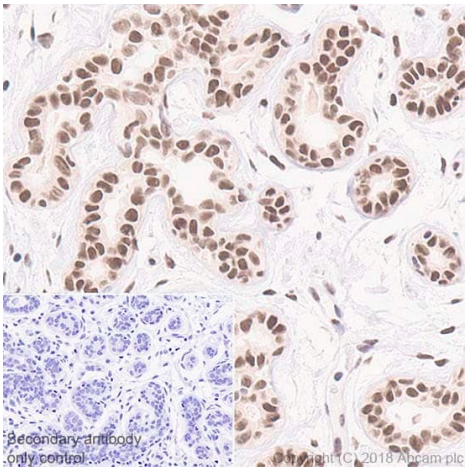
Exposure time: Lanes 1-3: 3 minutes; Lanes 4-6: 41 seconds.

The expression profile observed is consistent with what has been described in the literature (PMID: 26068589).



ChIP - Anti-Rad50 antibody [EPR20968] - ChIP Grade (ab208019)

Chromatin was prepared from HeLa (human epithelial cell line from cervix adenocarcinoma) cells treated with 1% methymethanesulfonate (MMS) according to the Abcam X-ChIP protocol. Cells were fixed with formaldehyde for 10min. The ChIP was performed with 25 µg of chromatin, 2 µg of ab208019 (red), and 20 µl of Protein A/G sepharose beads. 2µg of rabbit normal IgG was added to the beads control (gray). The immunoprecipitated DNA was quantified by real time PCR (SYBR green approach). rDNA primers used are located in the region of ribosomal gene loci following the publication PMID:21029860(rDNA 1: rDNA 13017F/13068R ; rDNA 2 : rDNA 1125F/1201R ; rDNA 3 : rDNA 30409F/30566R).

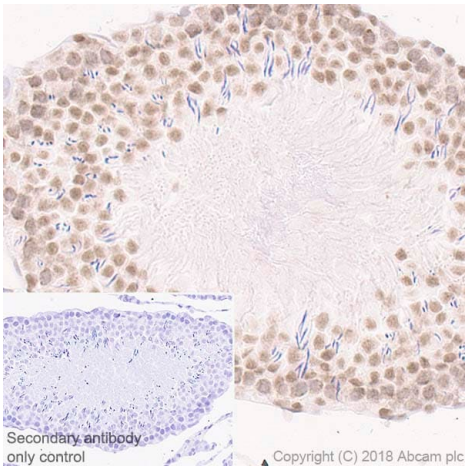


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Rad50 antibody [EPR20968] - ChIP Grade (ab208019)

Immunohistochemical analysis of paraffin-embedded human breast tissue labeling Rad50 with ab208019 at 1/4000 dilution, followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP). Nuclear staining in human breast tissue (PMID: 15509680) is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP).

Perform heat-mediated antigen retrieval using **ab93684** (Tris/EDTA buffer, pH 9.0).

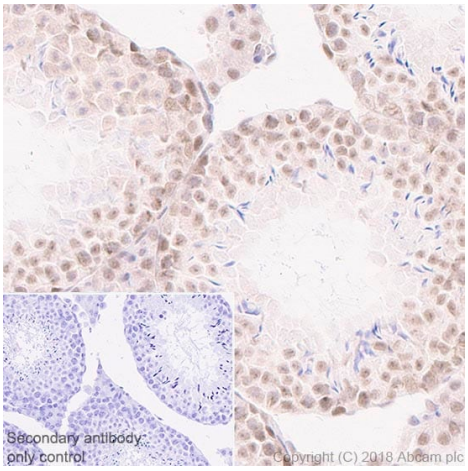


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Rad50 antibody [EPR20968] - ChIP Grade (ab208019)

Immunohistochemical analysis of paraffin-embedded rat testis tissue labeling Rad50 with ab208019 at 1/4000 dilution, followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP). Nuclear staining in rat testis is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP).

Perform heat-mediated antigen retrieval using **ab93684** (Tris/EDTA buffer, pH 9.0).

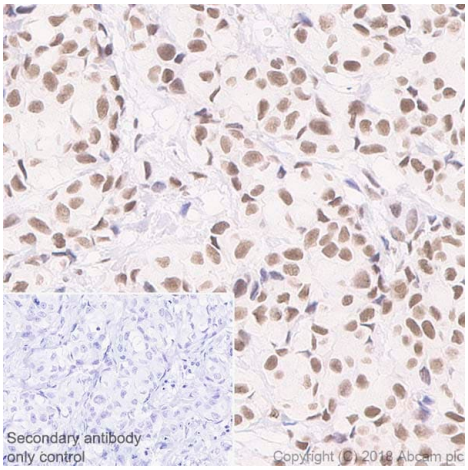


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Rad50 antibody [EPR20968] - ChIP Grade (ab208019)

Immunohistochemical analysis of paraffin-embedded mouse testis tissue labeling Rad50 with ab208019 at 1/4000 dilution, followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP). Nuclear staining in mouse testis (PMID: 10908350) is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP).

Perform heat-mediated antigen retrieval using **ab93684** (Tris/EDTA buffer, pH 9.0).

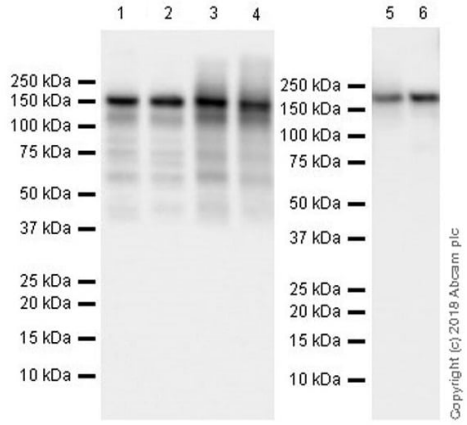


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Rad50 antibody [EPR20968] - ChIP Grade (ab208019)

Immunohistochemical analysis of paraffin-embedded human breast carcinoma tissue labeling Rad50 with ab208019 at 1/4000 dilution, followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP). Nuclear staining in human breast carcinoma (PMID: 24642965) is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP).

Perform heat-mediated antigen retrieval using **ab93684** (Tris/EDTA buffer, pH 9.0).



Western blot - Anti-Rad50 antibody [EPR20968] - ChIP Grade (ab208019)

All lanes : Anti-Rad50 antibody [EPR20968] - ChIP Grade (ab208019) at 1/1000 dilution

Lane 1 : K562 (human chronic myelogenous leukemia cell line from bone marrow) whole cell lysate
Lane 2 : HeLa (human epithelial cell line from cervix adenocarcinoma) whole cell lysate
Lane 3 : HEK-293T (human epithelial cell line from embryonic kidney transformed with large T antigen) whole cell lysate
Lane 4 : HT-29 (human colorectal adenocarcinoma cell line) whole cell lysate
Lane 5 : NIH/3T3 (Mouse embryo fibroblast cell line) whole cell lysate
Lane 6 : ES-D3 (mouse embryonic multipotent stem Cell) whole cell lysate

Lysates/proteins at 20 µg per lane.

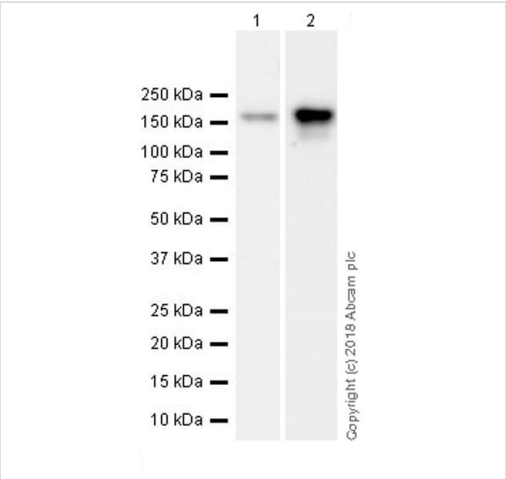
Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/100000 dilution

Predicted band size: 153 kDa

Blocking/Diluting buffer and concentration: 5% NFDM/TBST.

Exposure time: Lanes 1-4: 8 seconds; Lanes 5-6: 24 seconds.

The expression profile observed is consistent with what has been described in the literature (PMID: 26068589).



Western blot - Anti-Rad50 antibody [EPR20968] - ChIP Grade (ab208019)

All lanes : Anti-Rad50 antibody [EPR20968] - ChIP Grade (ab208019) at 1/1000 dilution

Lane 1 : Human fetal brain lysate

Lane 2 : Human fetal kidney lysate

Lysates/proteins at 10 µg per lane.

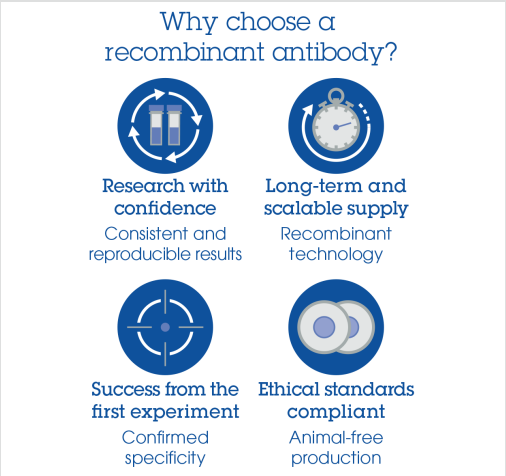
Secondary

All lanes : VeriBlot for IP Detection Reagent (HRP) (**ab131366**) at 1/4000 dilution

Predicted band size: 153 kDa

Blocking/Diluting buffer and concentration: 5% NFDM/TBST.

Exposure time: Line 1: 3 minutes; Line 2: 41 seconds.



Anti-Rad50 antibody [EPR20968] - ChIP Grade (ab208019)

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