abcam

ab254299

Anti-Eg5 antibody [EPR23277-60]

Recombinant | RabMAb

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	EPR23277-60
Purification technique	Affinity purification Protein A
Concentration	0.592 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IP

Tested

Species	Human
Dilution info	1/30
Notes	-

Not recommended

Species	Mouse, Rat
Dilution info	-
Notes	-

WB

Tested

Species	Human
Dilution info	1/1000
Notes	-

Not recommended

Species	Mouse, Rat
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	1/50
Notes	-

Not recommended

Species	Mouse, Rat
Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	1/500
Notes	-

Not recommended

Species	Mouse, Rat
Dilution info	-
Notes	-

IHC-P

Tested

Species	Human
Dilution info	1/500
Notes	-

Not recommended

Species	Mouse, Rat
Dilution info	-
Notes	-

Flow Cyt

Not recommended

Species	Rat, Mouse, Human
Dilution info	-
Notes	-

Target data

See full target information KIF11

Function	Motor protein required for establishing a bipolar spindle and thus contributing to chromosome congression during mitosis (PubMed:19001501, PubMed:37728657). Required in non-mitotic cells for transport of secretory proteins from the Golgi complex to the cell surface (PubMed:23857769).

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	Avoid freeze / thaw cycle

Notes

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.

Supplementary info

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

Activity summary	The Eg5 protein also known scientifically as kinesin family member 11 (KIF11) plays an important role in cell mitosis. It functions mechanically as a plus-end directed motor involved in the separation of centrosomes during the early stage of cell division. Eg5 with an approximate mass of 116 kDa is responsible for the proper alignment and segregation of spindle fibers. The protein is expressed in rapidly

	dividing cells particularly within tissues with high turnover rates such as bone marrow and epithelium.
Biological function summary	The motor function of Eg5 is indispensable for the mitotic spindle organization. It provides the force necessary to push microtubules in opposite directions. Eg5 functions as part of a complex cellular machinery that includes proteins like dynein and other kinesins. By controlling spindle bipolarity it ensures accurate chromosome segregation during cell division preventing errors that could lead to aneuploidy.
Pathways	The action of Eg5 is fundamental in mitotic pathways including the spindle assembly checkpoint pathway. It interacts with proteins such as cyclin-dependent kinases (CDKs) which regulate its activity to ensure proper cell cycle progression. The protein's role is integral in facilitating the dynamics of the metaphase spindle checkpoint guaranteeing cell cycle fidelity.
Associated diseases and disorders	Eg5 is linked to certain types of cancers where there is abnormal cell division. The overexpression or dysregulation of Eg5 can contribute to tumorigenesis. Additionally Eg5 interacts with proteins involved in cancer such as p53 a tumor suppressor protein directly influencing cellular proliferation and survival. Researchers target Eg5 in the development of anticancer therapies to inhibit its activity aiming to obstruct malignant cell proliferation.

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

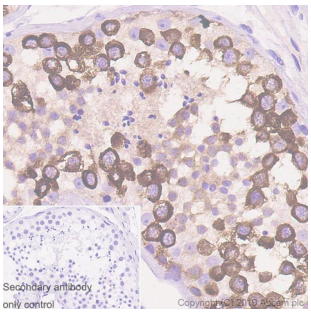
We do not recommend this combination. It is not covered by our product promise.

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.

8 product images

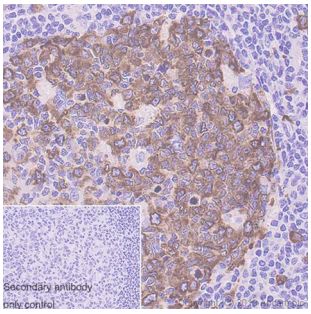


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Eg5 antibody [EPR23277-60] (ab254299)

Immunohistochemical analysis of paraffin-embedded Human testis tissue labeling Eg5 with ab254299 at 1/500 dilution (1.18 µg/ml) followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP) secondary antibody. Cytoplasmic staining in human tonsil (PMID:25277178) is observed.
The section was incubated with ab254299 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins.

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



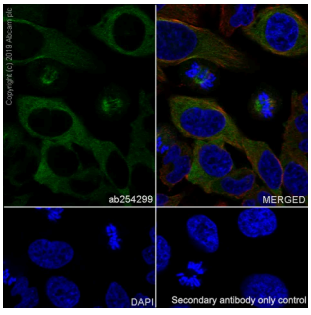
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Eg5 antibody [EPR23277-60] (ab254299)

Eg5 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-Eg5 antibody

Immunohistochemical analysis of paraffin-embedded Human tonsil tissue labeling Eg5 with ab254299 at 1/500 dilution, followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP) secondary antibody. Cytoplasmic staining in human tonsil (PMID:25277178) is observed.
The section was incubated with ab254299 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin.

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins.

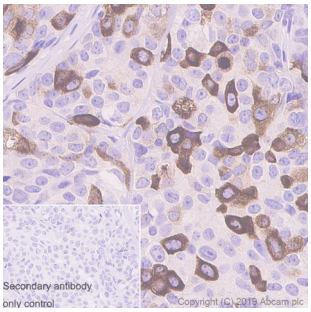
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP).



Immunocytochemistry/ Immunofluorescence - Anti-Eg5 antibody [EPR23277-60] (ab254299)

Eg5 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-Eg5 antibody

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (human cervix adenocarcinoma epithelial cell line) cells labelling Eg5 with ab254299 at 1/50 dilution, followed by ab150077 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) antibody at 1/1000 dilution (Green). Confocal image showing cytoplasmic staining in HeLa cells. ab195889 Anti-alpha Tubulin antibody (Alexa Fluor® 594) was used to counterstain tubulin at 1/100 dilution (Red). The Nuclear counterstain was DAPI (Blue).
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is ab150077 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) at 1/1000 dilution.



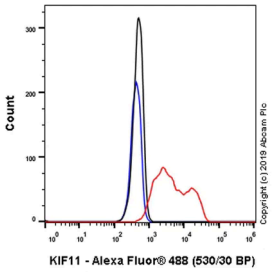
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Eg5 antibody [EPR23277-60] (ab254299)

Eg5 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-Eg5 antibody

Immunohistochemical analysis of paraffin-embedded Human breast cancer tissue labeling Eg5 with ab254299 at 1/500 dilution followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP) secondary antibody. Cytoplasmic staining in human tonsil (PMID:25277178) is observed. The section was incubated with ab254299 for 30 mins at room temperature.
The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin.

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins.

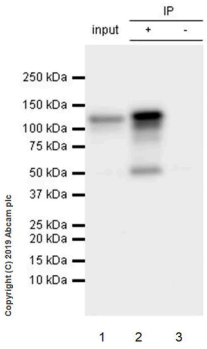
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP).



Flow Cytometry (Intracellular) - Anti-Eg5 antibody [EPR23277-60] (ab254299)

Eg5 Flow Cytometry (Intracellular) staining using rabbit Anti-Eg5 antibody

Intracellular flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized HeLa (Human cervix adenocarcinoma epithelial cell) cells labelling Eg5 with ab254299 at 1/500 dilution (Red) compared with a Rabbit monoclonal IgG (ab172730) isotype control (Black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue).
Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) at 1/2000 dilution was used as the secondary antibody.



Immunoprecipitation - Anti-Eg5 antibody [EPR23277-60] (ab254299)

Eg5 was immunoprecipitated from 0.35 mg HeLa (human cervix adenocarcinoma epithelial cell), whole cell lysate with ab254299 at 1/30 dilution. Western blot was performed on the immunoprecipitate using 254299 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (ab131366) was used at 1/5000 dilution.
Lane 1: HeLa whole cell lysate 10 µg

Lane 2: ab254299 IP in HeLa whole cell lysate

Lane 3: Rabbit monoclonal IgG (ab172730) instead of ab254299 in HeLa whole cell lysate.

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

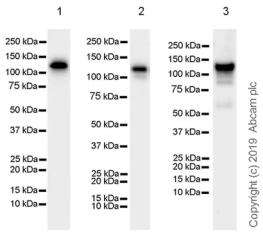
Exposure time: 3 seconds

The minor bands beneath the target band are probably caused by degradation.

All lanes:
Immunoprecipitation - Anti-Eg5 antibody [EPR23277-60] (ab254299)

Predicted band size: 119 kDa

Observed band size: 130 kDa



Western blot - Anti-Eg5 antibody [EPR23277-60] (ab254299)

Blocking and dilution buffer: 5% NFDM/TBST.
Exposure time:

Lanes 1-2: 15 seconds;
Lane 3: 5.5 seconds.

The expression profile observed is consistent with what has been described in the literature (PMID:23857769; 27492783).

Fresh lysates were tested in this WB.

All lanes:
Western blot - Anti-Eg5 antibody [EPR23277-60] (ab254299) at 1/1000 dilution

Lane 1:
HeLa (human cervix adenocarcinoma epithelial cell), whole cell lysate at 20 µg

Lane 2:
MCF7 (human breast adenocarcinoma epithelial cell), whole cell lysate at 20 µg

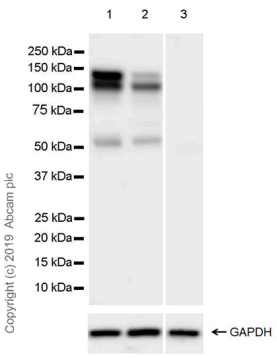
Lane 3:
Jurkat (human T cell leukemia T lymphocyte), whole cell lysate at 20 µg

Secondary

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

Predicted band size: 119 kDa

Observed band size: 130 kDa



Western blot - Anti-Eg5 antibody [EPR23277-60] (ab254299)

Blocking and dilution buffer: 5% NFDM/TBST.
Exposure time: 15 seconds

The expression profile observed is consistent with what has been described in the literature (PMID:23857769; 27492783).

Negative control: Human lung (PMID: 27279560).

The minor bands beneath the target band are probably caused by degradation.

All lanes:
Western blot - Anti-Eg5 antibody [EPR23277-60] (ab254299) at 1/1000 dilution

Lane 1:
Human testis tissue lysate at 20 µg

Lane 2:
Human tonsil tissue lysate at 20 µg

Lane 3:
Human lung tissue lysate at 20 µg

Secondary

All lanes:
Western blot - VeriBlot for IP Detection Reagent (HRP) (ab131366) at 1/1000 dilution

Predicted band size: 119 kDa

Observed band size: 130 kDa

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