abcam

Product datasheet

Anti-MAGEC2 antibody [EPR19064] ab209667

Recombinant RabMAb

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Overview

Product name Anti-MAGEC2 antibody [EPR19064]

Description Rabbit monoclonal [EPR19064] to MAGEC2

Host species Rabbit

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

Species reactivity Reacts with: Human, Recombinant fragment

Immunogen Recombinant full length protein. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: A-375 whole cell lysate; Full length recombinant protein MAGEE1. IHC-P: Human testis,

gastric cancer and melanoma tissues. ICC/IF: A-375 cells. Flow Cyt (intra): A-375 cells. IP: A-375

whole cell lysate.

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

Purity Protein A purified

Clonality Monoclonal EPR19064 Clone number

Isotype IgG

Applications

The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab209667 in the following tested applications.

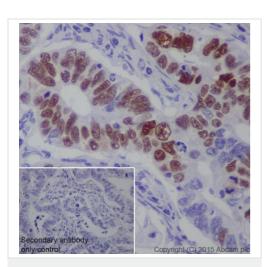
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
Flow Cyt (Intra)		1/150. ab172730 - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
WB		1/5000. Detects a band of approximately 51 kDa (predicted molecular weight: 41 kDa).
IHC-P		1/1000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
ICC/IF		1/500.
IP		1/40.

Target

Tissue specificity	Not expressed in normal tissues, except in germ cells in the seminiferous tubules and in Purkinje cells of the cerebellum. Expressed in various tumors, including melanoma, lymphoma, as well as pancreatic cancer, mammary gland cancer, non-small cell lung cancer and liver cancer. In hepatocellular carcinoma, there is a inverse correlation between tumor differentiation and protein expression, i.e. the lower the differentiation, the higher percentage of expression.	
Sequence similarities	Contains 1 MAGE domain.	
Developmental stage	Strongly expressed in spermatogonia and primary spermatocytes. At later stages of maturation, expression gradually decreases and becomes undetectable in mature spermatids.	
Cellular localization	Cytoplasm. Nucleus. Nuclear in germ cells. Cytoplasmic in well-differentiated hepatocellular carcinoma, nuclear in moderately- and poorly-differentiated hepatocellular carcinoma.	

Images

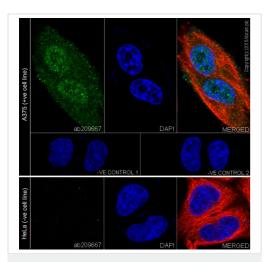


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-MAGEC2 antibody
[EPR19064] (ab209667)

Immunohistochemical analysis of paraffin-embedded Human gastric cancer tissue labeling MAGEC2 with ab209667 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution. Nucleus staining on Human gastric cancer is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/500 dilution.

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



Immunocytochemistry/ Immunofluorescence - Anti-MAGEC2 antibody [EPR19064] (ab209667)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized A-375 (Human malignant melanoma cell line) and HeLa (Human epithelial cell line from cervix adenocarcinoma) cells labeling MAGEC2 with ab209667 at 1/500 dilution, followed by Goat anti-rabbit lgG (Alexa Fluor[®] 488) (ab150077) secondary antibody at 1/1000 dilution (green).

Confocal image showing nuclear and weakly cytoplasmic staining on A-375 cell line

Negative expression in HeLa cells.

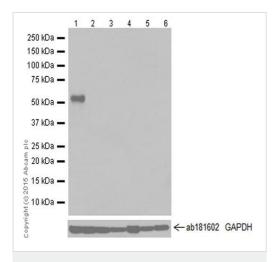
The nuclear counter stain is DAPI (blue).

Tubulin is detected with $\underline{ab7291}$ (anti-Tubulin mouse mAb) at 1/1000 dilution and $\underline{ab150120}$ (AlexaFluor $^{@}$ 594 Goat anti-Mouse secondary) at 1/1000 dilution (red).

The negative controls are as follows:

-ve control 1: ab209667 at 1/500 dilution followed by <u>ab150120</u> (AlexaFluor[®]594 Goat anti-Mouse secondary) at 1/1000 dilution.
-ve control 2: <u>ab7291</u> (anti-Tubulin mouse mAb) at 1/1000 dilution followed by <u>ab150077</u> (Alexa Fluor[®]488 Goat Anti-Rabbit IgG H&L)

at 1/1000 dilution.



Western blot - Anti-MAGEC2 antibody [EPR19064] (ab209667)

All lanes : Anti-MAGEC2 antibody [EPR19064] (ab209667) at 1/10000 dilution

Lane 1: A-375 (Human malignant melanoma cell line) whole cell lysate

Lane 2: HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate

Lane 3 : HepG2 (Human liver hepatocellular carcinoma cell line) whole cell lysate

Lane 4: Mouse testis lysate

Lane 5 : Human fetal kidney lysate

Lane 6 : Human fetal heart lysate

Lysates/proteins at 10 µg per lane.

Secondary

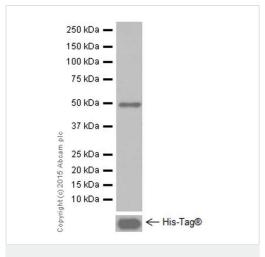
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Predicted band size: 41 kDa **Observed band size:** 51 kDa

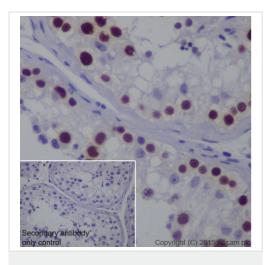
Exposure time: 30 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.

The band is consistent with what is observed in the literature (PMID: 16594646).



Western blot - Anti-MAGEC2 antibody [EPR19064] (ab209667)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-MAGEC2 antibody
[EPR19064] (ab209667)

Anti-MAGEC2 antibody [EPR19064] (ab209667) at 1/5000 dilution + Full length recombinant protein MAGEC2 at 0.01 µg

Secondary

Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Predicted band size: 41 kDa **Observed band size:** 51 kDa

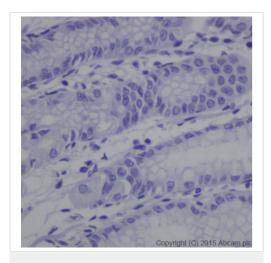
Exposure time: 1 second

Blocking/Dilution buffer: 5% NFDM/TBST.

Immunohistochemical analysis of paraffin-embedded Human testis tissue labeling MAGEC2 with ab209667 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution. Nucleus staining on Human germ cells is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/500 dilution.

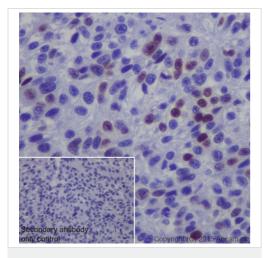
Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-MAGEC2 antibody
[EPR19064] (ab209667)

Immunohistochemical analysis of paraffin-embedded Human stomach tissue labeling MAGEC2 with ab209667 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution. Negative staining on Human stomach. Counter stained with Hematoxylin.

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

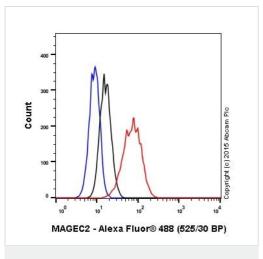


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-MAGEC2 antibody
[EPR19064] (ab209667)

Immunohistochemical analysis of paraffin-embedded Human melanoma tissue labeling MAGEC2 with ab209667 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution. Nucleus staining on melanoma is observed. Counter stained with Hematoxylin.

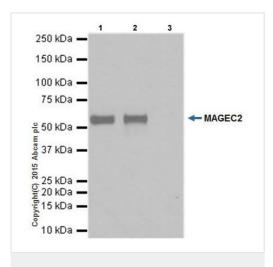
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/500 dilution.

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



Flow Cytometry (Intracellular) - Anti-MAGEC2 antibody [EPR19064] (ab209667)

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed A-375 (Human malignant melanoma cell line) cells labeling MAGEC2 with ab209667 at 1/150 dilution (red) compared with a rabbit monoclonal lgG isotype control (ab172730; black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody; blue). Goat anti rabbit lgG (Alexa Fluor® 488) at 1/500 dilution was used as the secondary antibody.



Immunoprecipitation - Anti-MAGEC2 antibody [EPR19064] (ab209667)

MAGEC2 was immunoprecipitated from 1mg of A-375 (Human malignant melanoma cell line) whole cell lysate with ab209667 at 1/40 dilution. Western blot was performed from the immunoprecipitate using ab209667 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/10000 dilution.

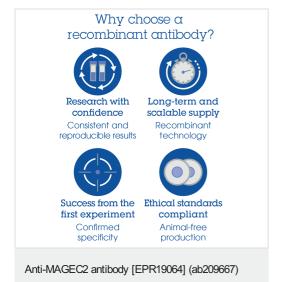
Lane 1: A-375 whole cell lysate 10µg (Input).

Lane 2: ab209667 IP in A-375 whole cell lysate.

Lane 3: Rabbit monoclonal IgG (<u>ab172730</u>) instead of ab209667 in A-375 whole cell lysate.

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

Exposure time: 1 second.



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