# abcam

## Product datasheet

# Anti-Geminin antibody [EPR14637] ab195047





重组 RabMAb

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概述

产品名称 Anti-Geminin抗体[EPR14637]

描述 兔单克隆抗体[EPR14637] to Geminin

宿主 Rabbit

经测试应用 适用于: WB, ICC/IF, IHC-P

种属反应性 与反应: Human

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

阳性对照 WB: HepG2, HeLa (treated with thymidine), and HeLa whole cell lysate. IHC-P: Human triple-

negative breast cancer and Human tonsil tissue. ICC/IF: MCF-7 cells

常规说明 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**.

性能

形式 Liquid

存放说明 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.

存储溶液 pH: 7.2

Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol, 0.05% BSA

纯度 Protein A purified

克隆 单克隆

克隆编号 EPR14637

同种型 ΙgG

#### The Abpromise guarantee

#### Abpromise™承诺保证使用ab195047于以下的经测试应用

"应用说明"部分下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

| 应用     | Ab评论             | 说明  |
|--------|------------------|---|
| WB     | <b>★★★★ (1)</b>  | 1/500. Predicted molecular weight: 24 kDa.  |
| ICC/IF | *** <u>*</u> (2) | 1/100.  |
| IHC-P  |                  | 1/500. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. |

#### 靶标

功能 Inhibits DNA replication by preventing the incorporation of MCM complex into prereplication

complex (pre-RC). It is degraded during the mitotic phase of the cell cycle. Its destruction at the

metaphase-anaphase transition permits replication in the succeeding cell cycle.

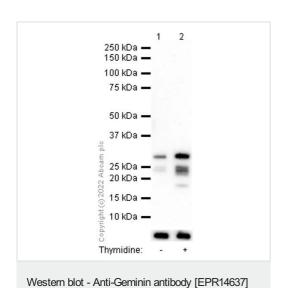
序列相似性 Belongs to the geminin family.

发展阶段 Absent during G1 phase, accumulates during S, G2, and M phases, and disappears at the time of

the metaphase-anaphase transition.

## 图片

(ab195047)



**All lanes :** Anti-Geminin antibody [EPR14637] (ab195047) at 1/1000 dilution

**Lane 1 :** Untreated HeLa (Human epithelial cells from cervix adenocarcinoma) whole cell lysate

Lane 2: HeLa treated with 2mM thymidine for 16 hours whole cell lysate

Lysates/proteins at 15 µg per lane.

#### **Secondary**

**All lanes :** Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

Predicted band size: 24 kDa

Exposure time: 20 seconds

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

Geminin is regulated during apoptosis by site-specific cleavage, resulting in the generation of two lower molecular weight cleavage products (21,462  $\pm$ 2.7 Da and 25,082  $\pm$ 2 Da). (PMID: 17261582)

260 kDa 160 kDa 125 kDa 70 kDa 38 kDa 30 kDa 25 kDa 15 kDa -

Western blot - Anti-Geminin antibody [EPR14637] (ab195047)

**All lanes :** Anti-Geminin antibody [EPR14637] (ab195047) at 1/500 dilution

Lane 1: Wild-type HeLa lysate

Lane 2: Geminin DNA Replication Inhibitor knockout HeLa lysate

Lane 3 : HepG2 lysate

Lane 4 : U-87 MG lysate

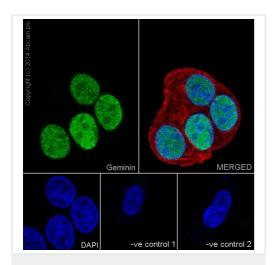
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 24 kDa

**Lanes 1-4:** Merged signal (red and green). Green - ab195047 observed at 32 kDa. Red - loading control <u>ab7291</u> observed at 50 kDa.

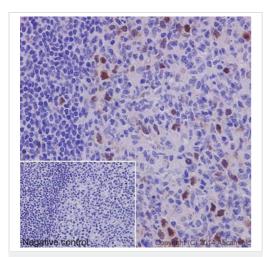
ab195047 Anti-Geminin antibody [EPR14637] was shown to specifically react with Geminin DNA Replication Inhibitor in wild-type HeLa cells. Loss of signal was observed when knockout cell line <a href="mailto:ab264972">ab264972</a> (knockout cell lysate <a href="mailto:ab257450">ab257450</a>) was used. Wild-type and Geminin DNA Replication Inhibitor knockout samples were subjected to SDS-PAGE. ab195047 and Anti-alpha Tubulin antibody [DM1A] - Loading Control (<a href="mailto:ab7291">ab7291</a>) were incubated overnight at 4°C at 1 in 500 and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (<a href="mailto:ab216773">ab216773</a>) and Goat anti-Mouse lgG H&L (IRDye® 680RD) preadsorbed (<a href="mailto:ab216776">ab216776</a>) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Immunocytochemistry/ Immunofluorescence - Anti-Geminin antibody [EPR14637] (ab195047)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized MCF-7 (Human breast adenocarcinoma cell line) cells labeling Geminin with ab195047 at 1/100 followed by Goat anti-rabbit lgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/500(green). Confocal image showing nuclear staining on MCF-7 cell line. The nuclear counterstain is DAPI (blue). Tubulin is detected with ab7291 (anti-Tubulin mouse mAb) at 1/1000 and ab150120 (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 (red).

The negative controls are as follows:-ve control 1 - ab195047 at 1/100 followed by **ab150120**(AlexaFluor®594 Goat anti-Mouse secondary) at 1/500.
-ve control 2. - **ab7291** (anti-Tubulin mouse mAb) at 1/1000 followed by **ab150077** (Alexa Fluor®488 Goat Anti-Rabbit IgG H&L) at 1/500.

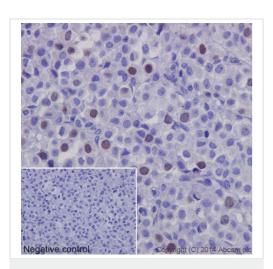


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Geminin antibody
[EPR14637] (ab195047)

Immunohistochemical analysis of paraffin-embedded Human tonsil tissue labeling Geminin with ab195047 at 1/500 followed by Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/500. Nucleus staining on Human tonsil tissue is observed. Counter stained with Hematoxylin.

Negative control: Used PBS instead of primary antibody and **ab97051** at 1/500 as secondary antibody.

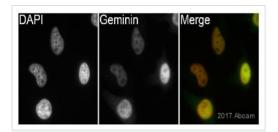
Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Geminin antibody
[EPR14637] (ab195047)

Immunohistochemical analysis of paraffin-embedded Human triplenegative breast cancer tissue labeling Geminin with ab195047 at 1/500 followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500. Nucleus staining on Human triple-negative breast cancer tissue is observed. Counter stained with Hematoxylin. Negative control: Used PBS instead of primary antibody and ab97051 at 1/500 as secondary antibody.

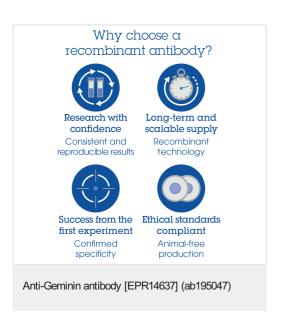
Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunocytochemistry/ Immunofluorescence - Anti-Geminin antibody [EPR14637] (ab195047)

This image is courtesy of an abreview by Kirk Mcmanus

ab195047 staining Geminin in the HeLa cell line from Human cervix by ICC/IF (Immunocytochemistry/immunofluorescence). Cells were fixed with paraformaldehyde, permeabilized with 0.5% Triton X-100 Samples were incubated with primary antibody (1/50 in PBS) for 1 hour at 22°C. Ab150081 (1/200) was used as the secondary antibody.



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