abcam

Product datasheet

Anti-cleaved N-terminal GSDMD antibody [EPR20829-408] ab215203

Recombinant RabMAb

**** 3 Abreviews 71 References 3 Images

Overview

Product name Anti-cleaved N-terminal GSDMD antibody [EPR20829-408]

Description Rabbit monoclonal [EPR20829-408] to cleaved N-terminal GSDMD

Host species Rabbit

Specificity The N-term cleaved Gasdermin-D needs inflammatory caspases in response to canonical and

non-canonical inflammasome activators (PubMed: 31548300, PubMed: 27418190, PubMed: 26375259). If need to detect cleavage of GSDMD, please ensure that the samples are treated

with inflammation before testing.

Tested applications Suitable for: WB

Unsuitable for: IHC-P

Species reactivity Reacts with: Human

Immunogen Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: Salmonella Infected (MOI:100) A431 whole cell lysate; THP-1 treated with 500 ng/ml Eprl for

2h, 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

1

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

Purity Protein A purified

Clonality Monoclonal

Clone number EPR20829-408

Isotype IgG

Applications

The Abpromise guarantee

Our Abpromise guarantee covers the use of ab215203 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)	1/1000. Detects a band of approximately 31 kDa (predicted molecular weight: 53 kDa).

Application notes

Is unsuitable for IHC-P.

Target

Function

Gasdermin-D, N-terminal: Promotes pyroptosis in response to microbial infection and danger signals. Produced by the cleavage of gasdermin-D by inflammatory caspases CASP1 or CASP4 in response to canonical, as well as non-canonical (such as cytosolic LPS) inflammasome activators (PubMed:26375003, PubMed:26375259, PubMed:27418190). After cleavage, moves to the plasma membrane where it strongly binds to inner leaflet lipids, including monophosphorylated phosphatidylinositols, such as phosphatidylinositol 4-phosphate, bisphosphorylated phosphatidylinositols, such as phosphatidylinositol (4,5)-bisphosphate, as well as phosphatidylinositol (3,4,5)-bisphosphate, and more weakly to phosphatidic acid and phosphatidylserine (PubMed:27281216). Homooligomerizes within the membrane and forms pores of 10 - 15 nanometers (nm) of inner diameter, possibly allowing the release of mature IL1B and triggering pyroptosis (PubMed:27418190, PubMed:27281216). Exhibits bactericidal activity. Gasdermin-D, N-terminal released from pyroptotic cells into the extracellular milieu rapidly binds to and kills both Gram-negative and Gram-positive bacteria, without harming neighboring mammalian cells, as it does not disrupt the plasma membrane from the outside due to lipidbinding specificity (PubMed:27281216). Under cell culture conditions, also active against intracellular bacteria, such as Listeria monocytogenes (By similarity). Strongly binds to bacterial and mitochondrial lipids, including cardiolipin. Does not bind to unphosphorylated phosphatidylinositol, phosphatidylethanolamine nor phosphatidylcholine (PubMed:27281216).

Tissue specificity

Expressed in the suprabasal cells of esophagus, as well as in the isthmus/neck, pit, and gland of the stomach, suggesting preferential expression in differentiating cells.

Sequence similarities

Belongs to the gasdermin family.

Domain

Intramolecular interactions between N- and C-terminal domains may be important for autoinhibition in the absence of cleavage by inflammatory caspases CASP1 or CASP4. The intrinsic pyroptosis-inducing activity is carried by gasdermin-D, N-terminal, that is released upon cleavage by inflammatory caspases.

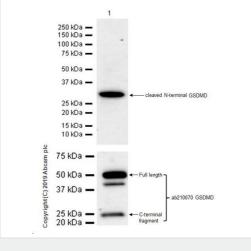
Post-translational modifications

Cleavage at Asp-275 by CASP1 (mature and uncleaved precursor forms) or CASP4 relieves autoinhibition and is sufficient to initiate pyroptosis.

Cellular localization

Cell membrane. Secreted. Released in the extracellular milieu following pyroptosis and Cytoplasm, cytosol. Inflammasome. In response to a canonical inflammasome stimulus, such as nigericin, recruited to NLRP3 inflammasone with similar kinetics to that of uncleaved CASP1 precursor.

Images



Western blot - Anti-cleaved N-terminal GSDMD antibody [EPR20829-408] (ab215203)

Anti-cleaved N-terminal GSDMD antibody [EPR20829-408] (ab215203) at 1/1000 dilution + Salmonella Infected (MOI:100) A431 (human epidermoid carcinoma cell line) whole cell lysate at 20 µg

Secondary

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

Predicted band size: 53 kDa **Observed band size:** 31 kDa

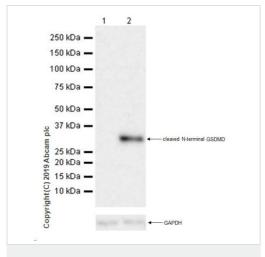
Exposure time: 3 minutes

The lysate was kindly provided by our collaborator Dr Feng Shao's lab, NIBS.

The molecular weight observed is consistent with literature (PMID: 26375003).

Blocking/Dilution buffer: 5% NFDM/TBST.

The N-term cleaved Gasdermin-D needs inflammatory caspases in response to canonical and non-canonical inflammasome activators (PubMed: 31548300, PubMed: 27418190, PubMed: 26375259). If need to detect cleavage of GSDMD, please ensure that the samples are treated with inflammation before testing.



Western blot - Anti-cleaved N-terminal GSDMD antibody [EPR20829-408] (ab215203)

All lanes : Anti-cleaved N-terminal GSDMD antibody [EPR20829-408] (ab215203) at 1/1000 dilution

Lane 1 : Untreated THP-1 (human epidermoid carcinoma cell line) whole cell lysate

Lane 2: THP-1 treated with 500 ng/ml Eprl for 2h, whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

Predicted band size: 53 kDa **Observed band size:** 31 kDa

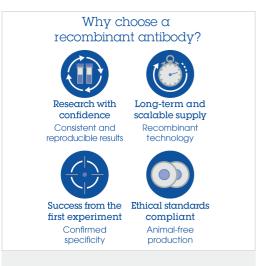
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Anti-cleaved N-terminal GSDMD antibody [EPR20829-408] (ab215203)

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