abcam

ab223877

Anti-DFNA5/GSDME antibody [EPR19859] - BSA and Azide free

Recombinant | RabMAb | KO Validated

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Constituents: PBS
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Carrier free	Yes
Clone number	EPR19859
Purification technique	Affinity purification Protein A
Specificity	Our previous testing data indicates that this antibody detects no signal or very weak signal in mouse heart, spleen, lung, stomach, testicle, skin, pancreas, muscle and lymph node tissues. Final results may be affected by the expression level of the tested samples.
Concentration	1.01 - 1.047 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IP

Tested

Species	Mouse
Dilution info	-
Notes	-

Expected

Species	Human, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

WB

Tested

Species	Human, Mouse, Rat
Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	-
Notes	-

Expected

Species	Mouse, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Target data

See full target information GSDME

Function	Gasdermin-E. Precursor of a pore-forming protein that converts non-inflammatory apoptosis to pyroptosis (PubMed:27281216, PubMed:28459430, PubMed:33852854, PubMed:35594856, PubMed:36607699). This form constitutes the precursor of the pore-forming protein: upon cleavage, the released N-terminal moiety (Gasdermin-E, N-terminal) binds to membranes and forms pores, triggering pyroptosis (PubMed:28459430). Gasdermin-E, N-terminal. Pore-forming protein produced by cleavage by CASP3 or granzyme B (GZMB), which converts non-inflammatory apoptosis to pyroptosis or promotes granzyme-mediated pyroptosis, respectively (PubMed:27281216, PubMed:28459430, PubMed:32188940, PubMed:33852854, PubMed:35594856). After cleavage, moves to the plasma membrane, homooligomerizes within the membrane and forms pores of 10-15 nanometers (nm) of inner diameter, allowing the release of mature interleukins (IL1B and IL16) and triggering pyroptosis (PubMed:28459430, PubMed:32188940, PubMed:33852854, PubMed:35594856). Binds to inner leaflet lipids, bisphosphorylated phosphatidylinositols, such as phosphatidylinositol (4,5)-bisphosphate (PubMed:28459430). Cleavage by CASP3 switches CASP3-mediated apoptosis induced by TNF or danger signals, such as chemotherapy drugs, to pyroptosis (PubMed:27281216, PubMed:28459430, PubMed:32188940). Mediates secondary necrosis downstream of the mitochondrial apoptotic pathway and CASP3 activation as well as in response to viral agents (PubMed:28045099). Exhibits bactericidal activity (PubMed:27281216). Cleavage by GZMB promotes tumor suppressor activity by triggering robust anti-tumor immunity (PubMed:21522185, PubMed:32188940). Suppresses tumors by mediating granzyme-mediated pyroptosis in target cells of natural killer (NK) cells: cleavage by granzyme B (GZMB), delivered to target cells from NK-cells, triggers pyroptosis of tumor cells and tumor suppression (PubMed:31953257, PubMed:32188940). May play a role in the p53/TP53-regulated cellular response to DNA damage (PubMed:16897187). Gasdermin-E, N-terminal. (Microbial infection) Pore-forming protein, which promotes maternal placental pyroptosis in response to Zika virus infection, contributing to adverse fetal outcomes.

Storage

Shipped at conditions	Blue Ice
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	+4°C
Storage information	Do Not Freeze

Notes

ab223877 is the carrier-free version of ab215191.

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.

Conjugation ready
Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with 1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

Compatibility
This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc.

Supplementary info

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

Activity summary	DFNA5 also known as GSDME (Gasdermin E) is a protein encoded by the GSDME gene. It has a molecular mass of approximately 59 kDa. The protein is expressed in various tissues including the cochlea in the inner ear and some epithelial tissues. Mechanically DFNA5/GSDME plays a role in inducing cell membrane pore formation leading to cell lysis. This activity relates to its involvement in processes like cell death specifically pyroptosis where the cell undergoes a form of programmed necrosis.
Biological function summary	DFNA5/GSDME serves as a pore-forming protein that facilitates pyroptotic cell death usually upon cleavage by caspase-3. This activity is important for innate immune response and the maintenance of cellular homeostasis. DFNA5/GSDME operates independently and does not form part of large protein complexes. The cleaved form inserts into the lipid bilayer of cell membranes contributing to the execution of cell death particularly under stress conditions or cellular insult.
Pathways	DFNA5/GSDME is involved in the pyroptosis and apoptosis pathways. It plays an essential role in the cellular response to inflammation and stress signals by interacting with caspase-3 a critical protease in the apoptosis pathway. The balance between apoptosis and pyroptosis decides cell fate with DFNA5/GSDME activation tipping towards pyroptotic cell death. The interconnection with other gasdermin family proteins such as GSDMD is notable as they share functional and structural similarities in promoting necrosis-like cell death processes.
Associated diseases and disorders	Mutations in the DFNA5 gene relate to progressive hearing loss (nonsyndromic sensorineural deafness). Dysregulation of GSDME has also been implicated in cancer where altered expression promotes tumorigenesis or tumor suppression depending on the context. In hearing loss GSDME's interaction with pathways influencing apoptosis highlights its pathogenic role while in cancer connections to caspase-3 show a potential dichotomy in cell survival and cell death regulation.

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.

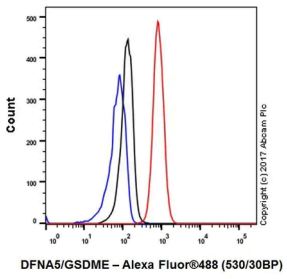
We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

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:
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6 product images

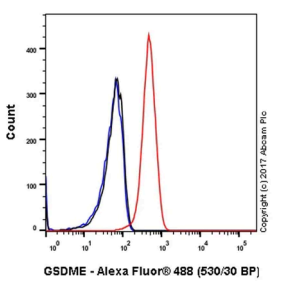


Flow Cytometry (Intracellular) - Anti-DFNA5/GSDME antibody [EPR19859] - BSA and Azide free (ab223877)

DFNA5/GSDME Flow Cytometry (Intracellular) staining using rabbit Anti-DFNA5/GSDME antibody

This Flow Cyt data was generated using the same anti-DFNA5/GSDME antibody clone [EPR19859] in a different buffer formulation (cat ab215191).

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed, 90% methanol permeabilized SH-SY5Y (human neuroblastoma cell line from bone marrow) cell line labeling DFNA5/GSDMEwith ab215191 at 1/60 dilution (red) compared with a Rabbit IgG, monoclonal [EPR25A] - Isotype Control (ab172730) (black) and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (blue). Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077) at 1/2000 dilution was used as the secondary antibody.



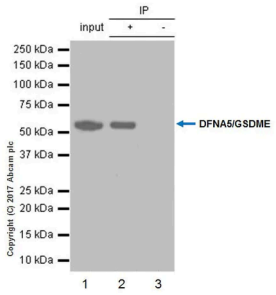
Flow Cytometry (Intracellular) - Anti-DFNA5/GSDME antibody [EPR19859] - BSA and Azide free (ab223877)

DFNA5/GSDME Flow Cytometry (Intracellular) staining using rabbit Anti-DFNA5/GSDME antibody

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed, 90% methanol permeabilized EMT6 cell line labeling DFNA5/GSDME with ab215191 at 1/600 dilution (red) compared with a Rabbit IgG, monoclonal [EPR25A] - Isotype Control (ab172730) (black) and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (blue). Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077) at 1/2000 dilution was used as the secondary antibody.

EMT6 cells were kindly provided by our collaborator Dr. Feng Shao, NIBS

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab215191).



Immunoprecipitation - Anti-DFNA5/GSDME antibody [EPR19859] - BSA and Azide free (ab223877)

ab215191 at 1/30 immunoprecipitating DFNA5/GSDME in EMT6 whole cell lysate.
Lane 1 (input): EMT6 whole cell lysate (10µg)

Lane 2 (+): ab215191 + EMT6 whole cell lysate

Lane 3 (-): Rabbit monoclonal IgG (ab172730) instead of ab215191 in EMT6 whole cell lysate

For western blotting, ab215191 at 1/500 and ab131366 VeriBlot for IP Detection Reagent (HRP) was used at 1/1000.

Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.

EMT6 cells were kindly provided by our collaborator Dr. Feng Shao, NIBS

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab215191).

All lanes:
Immunoprecipitation - Anti-DFNA5/GSDME antibody [EPR19859] - N-terminal (ab215191)

Predicted band size: 54 kDa

Exposure time: 55s

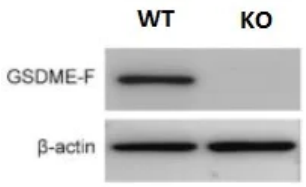


Image from Wang Y et al., Biochem Biophys Res Commu. 2018;495(1):1418-1425. Fig 4(A).; 10.1016/j.bbrc.2017.11.156 with permission from Elsevier.

Western blot - Anti-DFNA5/GSDME antibody [EPR19859] - BSA and Azide free (ab223877)

DFNA5/GSDME Western blot staining using rabbit Anti-DFNA5/GSDME antibody

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab215191).
https://www.sciencedirect.com/journal/biochemical-and-biophysical-research-communications

All lanes:
Western blot - Anti-DFNA5/GSDME antibody [EPR19859] - BSA and Azide free (ab223877) at 1/1000 dilution

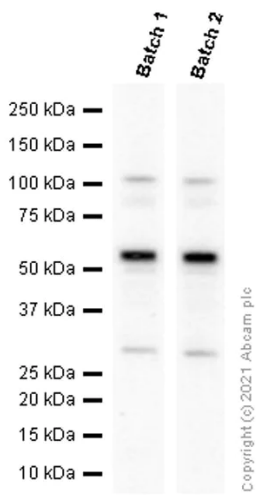
Lane 1:
Wild Type SGC-7901 cell lysates

Lane 2:
DFNA5/GSDME Knockout SGC-7901 cell lysates

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab6721) at 1/2000 dilution

Predicted band size: 54 kDa

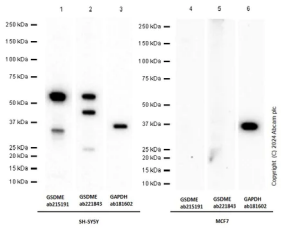


Western blot - Anti-DFNA5/GSDME antibody [EPR19859] - BSA and Azide free (ab223877)

DFNA5/GSDME Western blot staining using rabbit Anti-DFNA5/GSDME antibody

This data was developed using ab215191, the same antibody clone in a different buffer formulation. Different batches of ab215191 were tested on SH-SY5Y (Human neuroblastoma epithelial cell) lysate at 0.6 µg/ml. 15 µg of lysate was loaded in each lane. Bands observed at 55 kDa.
All lanes:
Western blot - Anti-DFNA5/GSDME antibody [EPR19859] - N-terminal (ab215191)

Predicted band size: 54 kDa



Western blot - Anti-DFNA5/GSDME antibody [EPR19859] - BSA and Azide free (ab223877)

DFNA5/GSDME Western blot staining using rabbit Anti-DFNA5/GSDME antibody

Blocking and dilution buffer:5% NFDM/TBST.
Exposure time: Lanes 1-3: 10 seconds , Lanes 4-6: 40 seconds.

Both ab215191 and ab221843 show negative result in MCF7, which is also consistent with the predicted expression pattern.

This data was developed using ab215191, the same antibody clone in a different buffer formulation.

Lanes 1 and 4:
Western blot - Anti-DFNA5/GSDME antibody [EPR19859] - N-terminal (ab215191) at 1/1000 dilution

Lanes 2 and 5:
Western blot - Anti-DFNA5/GSDME antibody [EPR19859-60] - C-terminal (ab221843) at 1/1000 dilution

Lanes 1 - 3:
SH-SY5Y (Human neuroblastoma epithelial cell) whole cell lysate at 20 µg

Lanes 4 - 6:
MCF7 (Human breast adenocarcinoma epithelial cell) whole cell lysate at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

Predicted band size: 54 kDa

Observed band size: 55 kDa

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