abcam

Product datasheet

Anti-GSDMD antibody [EPR20859] - BSA and Azide free ab239377

KO VALIDATED Recombinant RabMAb

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Overview

Product name	Anti-GSDMD antibody [EPR20859] - BSA and Azide free
Description	Rabbit monoclonal [EPR20859] to GSDMD - BSA and Azide free
Host species	Rabbit
Specificity	This antibody can detect full length, as well as a N-terminal fragment after stimulation in WB. Expression level of GSDMD in whole normal brain lysate is low or undetectable (PMID: 32671214, PMID: 34975487), we recommend loading higher amount of lysate or using lower antibody dilution.
Tested applications	**Suitable for:** Flow Cyt (Intra), IHC-P, WB, IP
Species reactivity	**Reacts with:** Mouse, Rat
Immunogen	Recombinant full length protein. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: Wild-type mouse liver and lung whole cell lysates; C6, PC-12, RAW 264.7 and NIH/3T3 whole cell lysates; Rat brain and liver lysates, IHC-P: Wild type mouse small intestine and mouse lung tissues. Flow Cyt (intra): RAW 264.7 cells. IP: RAW 264.7 whole cell lysate.
General notes	ab239377 is the carrier-free version of **ab219800**. Our **carrier-free** antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency. 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. 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. 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. Do Not Freeze.
Storage buffer	pH: 7.2 Constituent: PBS
Carrier free	Yes
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR20859
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab239377 in the following tested applications.

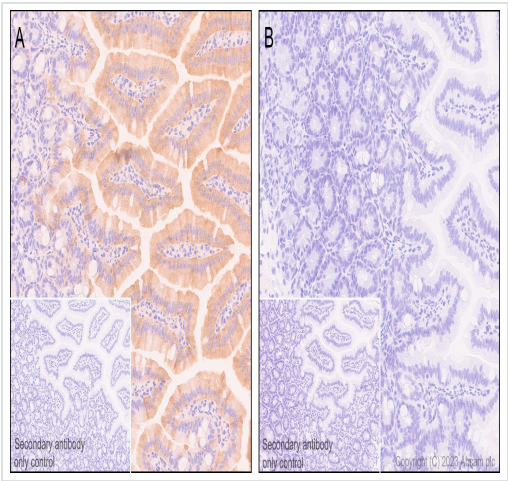
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
Flow Cyt (Intra)		Use at an assay dependent concentration.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
WB		Use at an assay dependent concentration. Detects a band of approximately 53 kDa (predicted molecular weight: 53 kDa).
IP		Use at an assay dependent concentration.

Target

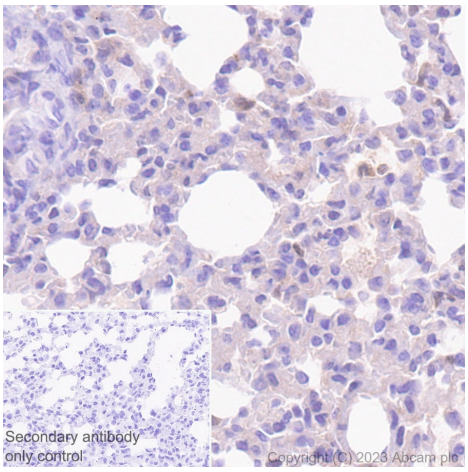
Sequence similarities Belongs to the gasdermin family.

Images



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-GSDMD antibody [EPR20859] - BSA and Azide free (ab239377)

This data was developed using **ab219800**, the same antibody clone in a different buffer formulation.
Immunohistochemical analysis of paraffin-embedded (A) Small intestine tissue from wild-type C57BL/6JGpt mice (B) Small intestine tissue from GSDMD knockout mice staining with **ab219800** at 1/5000 dilution and ready-to-use Goat Anti-Rabbit IgG H&L (HRP) secondary. Counterstaining with hematoxylin. Heat mediated antigen retrieval was performed with Citrate buffer (pH 6.0, Epitope Retrieval Solution 1) for 20 mins. Positive staining on (A) Small intestine tissue from wild-type C57BL/6JGpt mice and no staining on (B) Small intestine tissue from GSDMD knockout mice. The section was incubated with **ab219800** for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND™ RX instrument. The tissue samples were kindly provided by GemPharmatech. C57BL/6JGpt wildtype mice and GSDMD-KO homozygous mice (Strain ID: T010437).



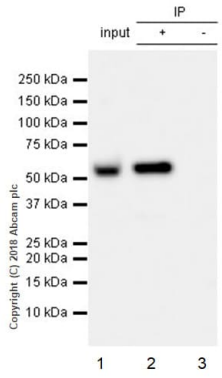
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-GSDMD antibody [EPR20859] - BSA and Azide free (ab239377)

Immunohistochemical analysis of paraffin-embedded mouse lung tissue labelling GSDMD with **ab219800** at 1/1000 (0.542 µg/ml) followed by **ab214880** at a ready to use dilution. **Low expression tissue:** weak staining on mouse lung. The section was incubated with **ab219800** at 4°C overnight. Counterstained with hematoxylin.

Secondary antibody only control: Secondary antibody is **ab214880** at a ready to use dilution.

Heat mediated antigen retrieval was performed using **ab93684** (Tris/EDTA buffer, pH 9.0).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab219800**).



Immunoprecipitation - Anti-GSDMD antibody [EPR20859] - BSA and Azide free (ab239377)

GSDMD was immunoprecipitated from 0.35 mg of RAW 264.7 (mouse macrophage cell line transformed with Abelson murine leukemia virus) whole cell lysate with **ab219800** at 1/30 dilution. Western blot was performed from the immunoprecipitate using **ab219800** at 1/500 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**), was used for detection at 1/1000 dilution.

Lane 1: RAW 264.7 whole cell lysate 10 µg (Input).

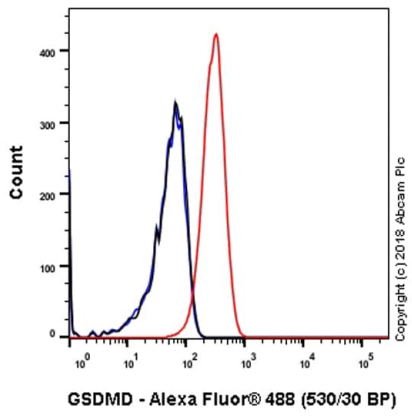
Lane 2: **ab219800** IP in RAW 264.7 whole cell lysate.

Lane 3: Rabbit monoclonal IgG (**ab172730**) instead of **ab219800** in RAW 264.7 whole cell lysate.

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

Exposure time: 10 seconds.

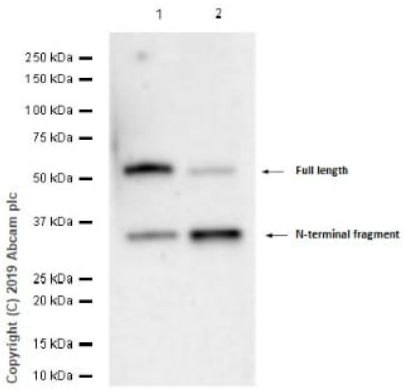
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab219800**).



Flow Cytometry (Intracellular) - Anti-GSDMD antibody [EPR20859] - BSA and Azide free (ab239377)

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed, 90% methanol-permeabilized RAW 264.7 (mouse macrophage cell line transformed with Abelson murine leukemia virus) cell line labeling GSDMD with **ab219800** at 1/500 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.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab219800**).



Western blot - Anti-GSDMD antibody [EPR20859] - BSA and Azide free (ab239377)

All lanes : Anti-GSDMD antibody [EPR20859] (**ab219800**) at 1/2000 dilution

Lane 1 : iBMM (mouse immortalized bone marrow derived macrophages) treated with 500ng/ml Bsak plus 500ng/ml anthrax protective antigen (PA) for 2h. Cell lysate
Lane 2 : iBMM (mouse immortalized bone marrow derived macrophages) treated with 500ng/ml Bsak plus 500ng/ml anthrax protective antigen (PA) for 2h. Concentrated cell supernatant

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

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

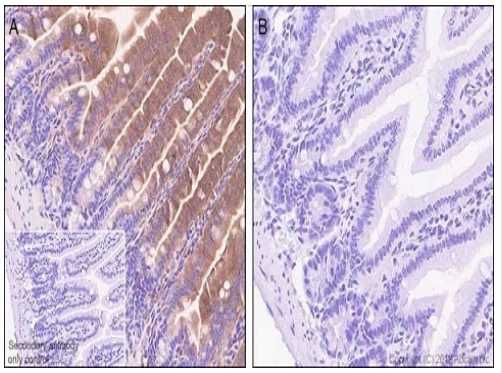
Exposure time: 180 seconds

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab219800**).

Blocking and diluting buffer: 5% NFDM /TBST

This antibody could detect full length, as well as a N-terminal fragment after stimulation.

The tissue samples were kindly provided by Dr Feng Shao's lab, NIBS



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-GSDMD antibody [EPR20859] - BSA and Azide free (ab239377)

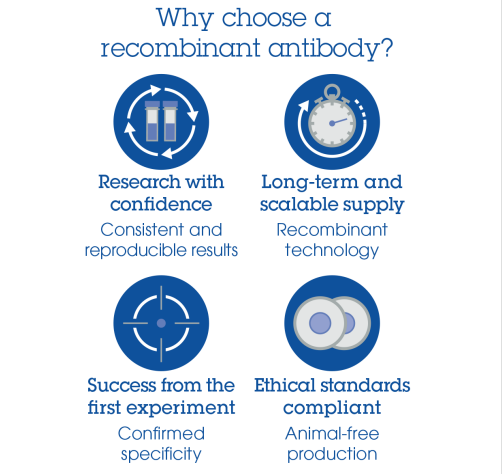
Immunohistochemical analysis of paraffin-embedded Wild type mouse small intestine (A) and GSDMD KO mouse small intestine (B) tissue labeling GSDMD with **ab219800** at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Cytoplasmic staining on epithelium of wild type mouse small intestine (A), no staining on GSDMD KO mouse small intestine (B) (PMID: 17350798) is observed. Counter stained with hematoxylin.

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

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab219800**).

Heat mediated antigen retrieval was performed with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

The tissue samples were kindly provided by Dr Feng Shao's lab, NIBS.



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