abcam

ab310335

Anti-Neutrophil Elastase antibody [EPR28386-66]

Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335) is a rabbit monoclonal antibody detecting Neutrophil Elastase in **Western Blot, Flow Cytometry (Intra), IHC-P, mIHC**. Suitable for **Human, Mouse, Rat**.

- Multiplex IHC validated on the Leica BOND® MAX using Opal reagents
- Biophysical QC for unrivalled batch-batch consistency

Recombinant | RabMAb | Advanced Validation | 20ul selling size

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Clone number	EPR28386-66
Purification technique	Affinity purification Protein A
Concentration	0.51 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

WB

Tested

Species	Human
Dilution info	1/1000
Notes	ab310335 is not suitable for detecting human cell lines in western blot assay due to low sensitivity issue. ab131260 and ab314916 are alternatives for detecting human cell lines.

Species	Mouse
Dilution info	1/1000
Notes	0.538 ug/ml

Species	Rat
Dilution info	1/1000
Notes	0.538 ug/ml

Flow Cyt (Intra)

Tested

Species	Mouse
Dilution info	1/500
Notes	-

Expected

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

IHC-P

Tested

Species	Human
Dilution info	1/100
Notes	Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Species	Mouse
Dilution info	1/100
Notes	Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Species	Rat
Dilution info	1/100
Notes	Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

ICC/IF

Not recommended

Species	Mouse, Human, Rat
Dilution info	-
Notes	-

IP

Not recommended

Species	Mouse, Rat, Human
Dilution info	-
Notes	-

mIHC

Tested

Species	Human
Dilution info	1/100
Notes	Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Species	Mouse
Dilution info	1/500

Notes	-

Expected

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

Target data

See full target information Elane

Function	Serine protease that modifies the functions of natural killer cells, monocytes and granulocytes. Inhibits C5a-dependent neutrophil enzyme release and chemotaxis (By similarity). Promotes blood coagulation (PubMed:20676107). Through the activation of the platelet fibrinogen receptor integrin alpha-IIb/beta-3, potentiates platelet aggregation induced by a threshold concentration of cathepsin G (CTSG) (By similarity). Cleaves and thus inactivates tissue factor pathway inhibitor (TFPI) (By similarity). Capable of killing E.coli; probably digests outer membrane protein A (ompA) in E.coli (PubMed:10947984).

Storage

Shipped at conditions	Conditional Ambient
Appropriate short-term storage duration	1-2 weeks
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	-20°C
Aliquoting information	Upon delivery aliquot
Storage information	Avoid freeze / thaw cycle

Notes

What is this antibody validated in?
Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335) is a rabbit recombinant monoclonal antibody and is validated for use in Western Blot (WB), Flow Cytometry (Intra), Flow Cytometry (Flow Cyt), Immunohistochemistry (IHC-P), Multiplex IHC (mIHC) in Human, Mouse, Rat samples.

What is the molecular weight of Neutrophil Elastase?
Anti-Neutrophil Elastase [EPR28386-66] (ab310335) specifically detects a band for Neutrophil Elastase (UniProt: Q3UP87) at a molecular weight of 29kDa.

Other related products
We have a range of other formats of antibody clone [EPR28386-66] also available for your convenience: ab310335, Carrier free - ab310336

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.

What are the advantages of a recombinant monoclonal antibody?
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 batch production

For more information, read more on recombinant antibodies.

Trial sizes available!
Test your antibody or perform pre-screening before committing to a larger quantity. Sold in 20µl. Discover our selection of trial-size antibodies.

Supplementary info

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

Activity summary	Neutrophil Elastase also known as neutrophil proteinase 3 has a molecular mass of approximately 30 kDa. It is a serine protease expressed mainly in azurophilic granules of neutrophils. This enzyme breaks down various proteins including elastin at sites of inflammation. Neutrophil Elastase releases during degranulation or through neutrophil extracellular traps (NETs) to combat pathogens.
Biological function summary	The target plays a significant role in neutrophil-mediated tissue remodeling and host defense. It leads to the breaking down of extracellular matrix components contributing to tissue remodeling and repair. Neutrophil Elastase is part of a complex interplay with other proteases like matrix metalloproteinases (MMPs). Its activity is regulated by endogenous inhibitors such as alpha-1 antitrypsin to prevent excessive tissue damage.
Pathways	This protease is important in inflammation and immune response pathways. It plays a role in the complement system by activating C5 a component of the complement cascade. It also acts in conjunction with other proteases such as cathepsin G to modulate immune and inflammatory responses. Neutrophil Elastase's role within these pathways is pivotal for the regulation of inflammation and destruction of pathogens.

Associated diseases and disorders	Individuals with conditions such as chronic obstructive pulmonary disease (COPD) and cystic fibrosis often exhibit dysregulation or excessive activity of Neutrophil Elastase. This excessive activity leads to tissue damage and exacerbates disease progression. The enzyme also interacts with proteins like elastin and collagen in these diseases further contributing to pathological tissue remodeling and damage.

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

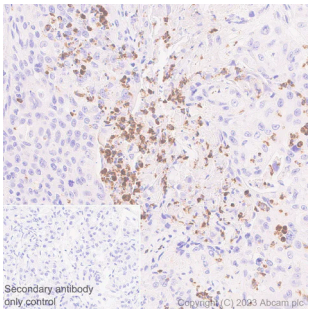
We do not recommend this combination. It is not covered by our product promise.

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.

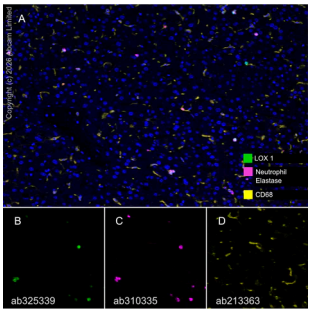
Full details and terms and conditions can be found here:
Terms & Conditions.

25 product images



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335)

Immunohistochemical analysis of paraffin-embedded Human cervical cance tissue labeling Neutrophil Elastase with ab310335 at 1/100 (5.38 ug/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining on neutrophils in human cervical cancer. The section was incubated with ab310335 for 30 mins at room temperature.The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin. Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins



Multiplex immunohistochemistry - Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335)

Neutrophil Elastase Multiplex immunohistochemistry staining of Human liver using rabbit Anti-Neutrophil Elastase antibody

Multiplex immunohistochemistry analysis of formalin/PFA-fixed paraffin-embedded human liver tissue staining LOX 1 with ab325339 at a 1/100 (5.05 µg/ml) dilution, ab310335 anti-Neutrophil Elastase used at a 1/100 (5.20 µg/ml) dilution and ab213363 anti-CD68 used at a 1/500 (1.314 µg/ml) dilution.
Panel A: anti-LOX 1 (green; Opal™520), anti-Neutrophil Elastase (magenta; Opal™570), anti-CD68 (yellow; Opal™690) on human liver.

Panel B: anti-LOX 1 staining neutrophils in human liver.

Panel C: anti-Neutrophil Elastase staining neutrophils in human liver.

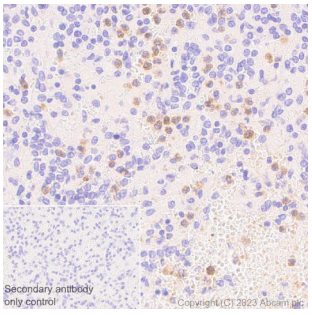
Panel D: anti-CD68 staining on macrophages in human liver.

Nuclear DNA was labeled with DAPI (shown in blue).

The section was incubated in three rounds of staining: in the order of ab325339, ab310335 and ab213363 for 30 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system.

The immunostaining was performed on a Leica Biosystems BOND® RX instrument with an Opal™ 4-color kit. Image acquisition was performed with Leica SP8 confocal microscope.

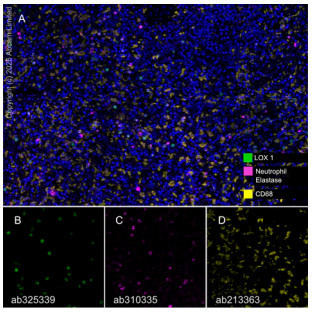
Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335)

Immunohistochemical analysis of paraffin-embedded Human spleen tissue labeling Neutrophil Elastase with ab310335 at 1/100 (5.38 ug/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining on neutrophils in human spleen. The section was incubated with ab310335 for 30 mins at room temperature.The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin. Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins

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Multiplex immunohistochemistry - Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335)

Neutrophil Elastase Multiplex immunohistochemistry staining of Human spleen using rabbit Anti-Neutrophil Elastase antibody

Multiplex immunohistochemistry analysis of formalin/PFA-fixed paraffin-embedded human spleen tissue staining LOX 1 with ab325339 at a 1/100 (5.05 µg/ml) dilution, ab310335 anti-Neutrophil

Elastase used at a 1/100 (5.20 µg/ml) dilution and ab213363 anti-CD68 used at a 1/500 (1.314 µg/ml) dilution.
Panel A: anti-LOX 1 (green; Opal™520), anti-Neutrophil Elastase (magenta; Opal™570), anti-CD68 (yellow; Opal™690) on human spleen.

Panel B: anti-LOX 1 staining neutrophils in human spleen.

Panel C: anti-Neutrophil Elastase staining neutrophils in human spleen.

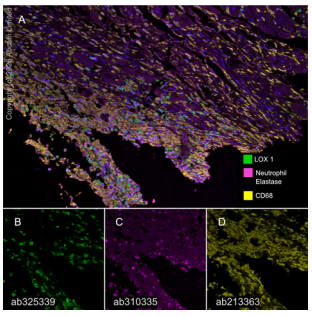
Panel D: anti-CD68 staining on macrophages in human spleen.

Nuclear DNA was labeled with DAPI (shown in blue).

The section was incubated in three rounds of staining: in the order of ab325339, ab310335 and ab213363 for 30 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system.

The immunostaining was performed on a Leica Biosystems BOND® RX instrument with an Opal™ 4-color kit. Image acquisition was performed with Leica SP8 confocal microscope.

Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins.



Multiplex immunohistochemistry - Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335)

Neutrophil Elastase Multiplex immunohistochemistry staining of Human heart atherosclerosis using rabbit Anti-Neutrophil Elastase antibody

Multiplex immunohistochemistry analysis of formalin/PFA-fixed paraffin-embedded human heart atherosclerosis tissue staining LOX 1 with ab325339 at a 1/100 (5.05 µg/ml) dilution, ab310335 anti-Neutrophil Elastase used at a 1/100 (5.20 µg/ml) dilution and ab213363 anti-CD68 used at a 1/500 (1.314 µg/ml) dilution.
Panel A: anti-LOX 1 (green; Opal™520), anti-Neutrophil Elastase (magenta; Opal™570), anti-CD68 (yellow; Opal™690) on human heart atherosclerosis.

Panel B: anti-LOX 1 staining neutrophils in human heart atherosclerosis.

Panel C: anti-Neutrophil Elastase staining neutrophils in human heart atherosclerosis.

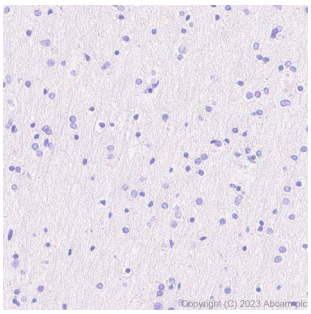
Panel D: anti-CD68 staining on macrophages in human heart atherosclerosis.

Nuclear DNA was labeled with DAPI (shown in blue).

The section was incubated in three rounds of staining: in the order of ab325339, ab310335 and ab213363 for 30 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system.

The immunostaining was performed on a Leica Biosystems BOND® RX instrument with an Opal™ 4-color kit. Image acquisition was performed with Leica SP8 confocal microscope.

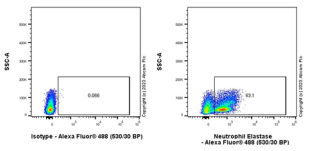
Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335)

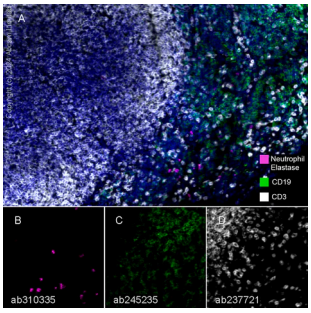
Neutrophil Elastase Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of Human cerebrum using rabbit Anti-Neutrophil Elastase antibody

Immunohistochemical analysis of paraffin-embedded Human cerebrum tissue labeling Neutrophil Elastase with ab310335 at 1/100 (5.38 ug/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Negative control: No staining on human cerebrum. The section was incubated with ab310335 for 30 mins at room temperature.The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin. Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins



Flow Cytometry (Intracellular) - Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335)

Flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized Mouse bone marrow cells labelling Neutrophil Elastase with ab310335 at 1/500 dilution (0.1 ug)/Right (Red) compared with a Rabbit monoclonal IgG (ab172730) / Left isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat Anti-Rabbit IgG (Alexa Fluor® 488, ab150081) at 1/5000 dilution was used as the secondary antibody.



Multiplex immunohistochemistry - Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335)

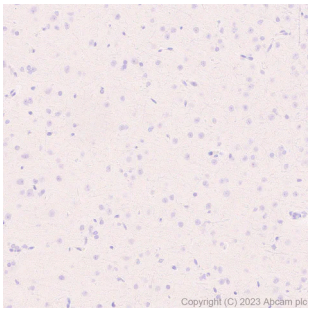
Multiplex immunohistochemistry analysis of formalin/PFA-fixed paraffin-embedded Mouse lymph node tissue staining Neutrophil Elastase with ab310335 at a 1/500 (1.076 µg/ml) dilution, CD19 with ab245235 at a 1/1000 (0.444 µg/ml) dilution and CD3 with ab237721 at a 1/2000 (0.264 µg/ml) dilution followed by a ready to use secondary antibody Opal Polymer HRP Ms + Rb.

Panel A: merged staining of anti-Neutrophil Elastase (magenta; Opal™690), anti-CD19 (green; Opal™520) and anti-CD3 (gray; Opal™690) on mouse spleen.Panel B: anti-Neutrophil Elastase staining neutrophils in mouse spleen.
Panel C: anti-CD19 staining B lymphocytes in mouse spleen.
Panel D: anti-CD3 staining T lymphocytes in mouse spleen.
Nuclear DNA was labeled with DAPI (shown in blue).

The section was incubated in three rounds of staining: in the order of ab310335, ab245235 and ab237721 for 30 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system.

The immunostaining was performed on a Leica Biosystems BOND® RX instrument with an Opal™ 4-color kit. Image acquisition was performed with Leica SP8 confocal microscope.

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins

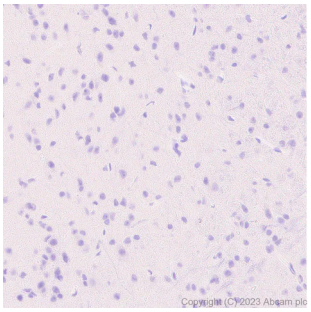


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335)

Neutrophil Elastase Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of Rat cerebrum using rabbit Anti-Neutrophil Elastase antibody

Immunohistochemical analysis of paraffin-embedded Rat cerebrum tissue labeling Neutrophil Elastase with ab310335 at 1/100 (5.38 ug/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Negative control: No staining on rat cerebrum. The section was incubated with ab310335 for 30 mins at room temperature.The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin. Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins

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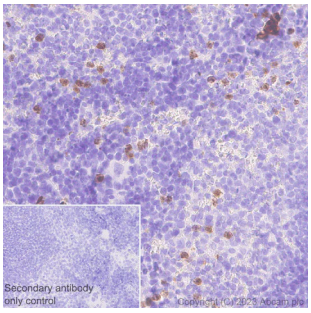


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335)

Neutrophil Elastase Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of Mouse cerebrum using rabbit Anti-Neutrophil Elastase antibody

Immunohistochemical analysis of paraffin-embedded Mouse cerebrum tissue labeling Neutrophil Elastase with ab310335 at 1/500 (1.076 ug/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Negative control: No staining on mouse cerebrum. The section was incubated with ab310335 for 30 mins at room temperature.The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin. Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins

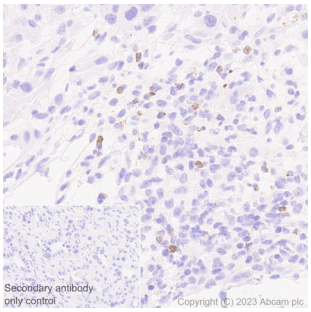
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Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335)

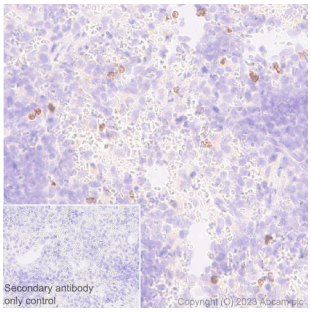
Immunohistochemical analysis of paraffin-embedded Mouse spleen tissue labeling Neutrophil Elastase with ab310335 at 1/500 (1.076 ug/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining on neutrophils in mouse spleen. The section was incubated with ab310335 for 30 mins at room temperature.The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin. Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins

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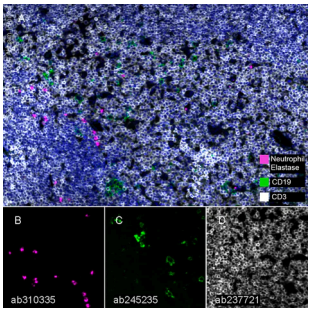
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335)

Immunohistochemical analysis of paraffin-embedded Mouse pancreatic can tissue labeling Neutrophil Elastase with ab310335 at 1/500 (1.076 ug/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining on neutrophils in mouse pancreatic cancer. The section was incubated with ab310335 for 30 mins at room temperature.The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin. Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335)

Immunohistochemical analysis of paraffin-embedded Rat spleen tissue labeling Neutrophil Elastase with ab310335 at 1/100 (5.38 ug/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining on neutrophils in rat spleen. The section was incubated with ab310335 for 30 mins at room temperature.The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin. Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins



Multiplex immunohistochemistry - Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335)

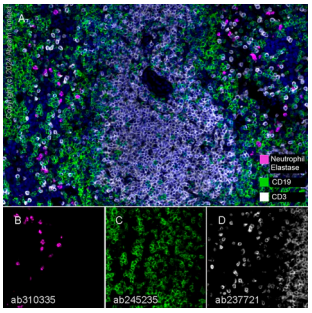
Multiplex immunohistochemistry analysis of formalin/PFA-fixed paraffin-embedded Mouse thymus tissue staining Neutrophil Elastase with ab310335 at a 1/500 (1.076 µg/ml) dilution, CD19 with ab245235 at a 1/1000 (0.444 µg/ml) dilution and CD3 with ab237721 at a 1/2000 (0.264 µg/ml) dilution followed by a ready to use secondary antibody Opal Polymer HRP Ms + Rb.

Panel A: merged staining of anti-Neutrophil Elastase (magenta; Opal™690), anti-CD19 (green; Opal™520) and anti-CD3 (gray; Opal™690) on mouse spleen.Panel B: anti-Neutrophil Elastase staining neutrophils in mouse spleen.
Panel C: anti-CD19 staining B lymphocytes in mouse spleen.
Panel D: anti-CD3 staining T lymphocytes in mouse spleen.
Nuclear DNA was labeled with DAPI (shown in blue).

The section was incubated in three rounds of staining: in the order of ab310335, ab245235 and ab237721 for 30 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system.

The immunostaining was performed on a Leica Biosystems BOND® RX instrument with an Opal™ 4-color kit. Image acquisition was performed with Leica SP8 confocal microscope.

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins



Multiplex immunohistochemistry - Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335)

Multiplex immunohistochemistry analysis of formalin/PFA-fixed paraffin-embedded Mouse spleen tissue staining Neutrophil Elastase with ab310335 at a 1/500 (1.076 µg/ml) dilution, CD19 with ab245235 at a 1/1000 (0.444 µg/ml) dilution and CD3 with ab237721 at a 1/2000 (0.264 µg/ml) dilution followed by a ready to use secondary antibody Opal Polymer HRP Ms + Rb.

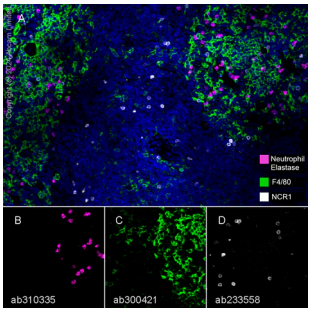
Panel A: merged staining of anti-Neutrophil Elastase (magenta; Opal™690), anti-CD19 (green; Opal™520) and anti-CD3 (gray; Opal™690) on mouse spleen.Panel B: anti-Neutrophil Elastase staining neutrophils in mouse spleen.
Panel C: anti-CD19 staining B lymphocytes in mouse spleen.
Panel D: anti-CD3 staining T lymphocytes in mouse spleen.
Nuclear DNA was labeled with DAPI (shown in blue).

The section was incubated in three rounds of staining: in the order of ab310335, ab245235 and ab237721 for 30 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system.

The immunostaining was performed on a Leica Biosystems BOND® RX instrument with an Opal™ 4-color kit. Image acquisition was performed with Leica SP8 confocal microscope.

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins

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Multiplex immunohistochemistry - Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335)

Neutrophil Elastase Multiplex immunohistochemistry staining of Mouse spleen using rabbit Anti-Neutrophil Elastase antibody

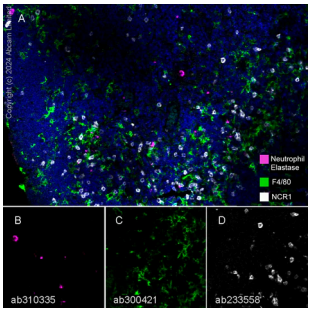
Multiplex immunohistochemistry analysis of formalin/PFA-fixed paraffin-embedded Mouse spleen tissue staining Neutrophil Elastase with ab310335 at a 1/500 (1.076 µg/ml) dilution, F4/80 with ab300421 at a 1/5000 (0.106 µg/ml) dilution and NCR1 with ab233558 at a 1/500 (1.114 µg/ml) dilution followed by a ready to use secondary antibody Opal Polymer HRP Ms + Rb.

Panel A: merged staining of anti-Neutrophil Elastase (magenta; Opal™690), anti-CD19 (green; Opal™520) and anti-CD3 (gray; Opal™690) on mouse spleen.Panel B: anti-Neutrophil Elastase staining neutrophils in mouse spleen.
Panel C: anti-CD19 staining B lymphocytes in mouse spleen.
Panel D: anti-CD3 staining T lymphocytes in mouse spleen.
Nuclear DNA was labeled with DAPI (shown in blue).

The section was incubated in three rounds of staining: in the order of ab310335, ab245235 and ab237721 for 30 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system.

The immunostaining was performed on a Leica Biosystems BOND® RX instrument with an Opal™ 4-color kit. Image acquisition was performed with Leica SP8 confocal microscope.

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins



Multiplex immunohistochemistry - Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335)

Neutrophil Elastase Multiplex immunohistochemistry staining of Mouse thymus using rabbit Anti-Neutrophil Elastase antibody

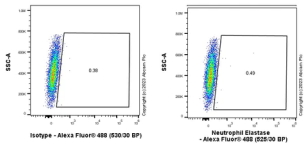
Multiplex immunohistochemistry analysis of formalin/PFA-fixed paraffin-embedded Mouse thymus tissue staining Neutrophil Elastase with ab310335 at a 1/500 (1.076 µg/ml) dilution, F4/80 with ab300421 at a 1/5000 (0.106 µg/ml) dilution and NCR1 with ab233558 at a 1/500 (1.114 µg/ml) dilution followed by a ready to use secondary antibody Opal Polymer HRP Ms + Rb.

Panel A: merged staining of anti-Neutrophil Elastase (magenta; Opal™690), anti-CD19 (green; Opal™520) and anti-CD3 (gray; Opal™690) on mouse thymus.
Panel B: anti-Neutrophil Elastase staining neutrophils in mouse thymus.
Panel C: anti-CD19 staining B lymphocytes in mouse thymus.
Panel D: anti-CD3 staining T lymphocytes in mouse thymus.
Nuclear DNA was labeled with DAPI (shown in blue).

The section was incubated in three rounds of staining: in the order of ab310335, ab245235 and ab237721 for 30 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system.

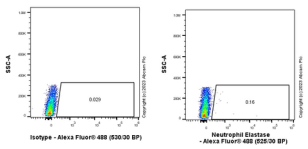
The immunostaining was performed on a Leica Biosystems BOND® RX instrument with an Opal™ 4-color kit. Image acquisition was performed with Leica SP8 confocal microscope.

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins



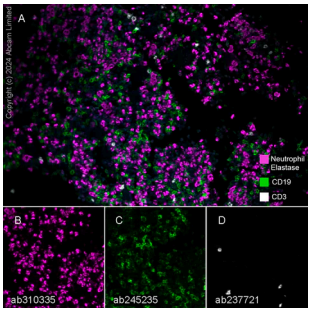
Flow Cytometry (Intracellular) - Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335)

Flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized NIH/3T3 (mouse embryonic fibroblast) cells labelling Neutrophil Elastase with ab310335 at 1/500 dilution (0.1 ug)/Right (Red) compared with a Rabbit monoclonal IgG (ab172730) / Left isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat Anti-Rabbit IgG (Alexa Fluor® 488, ab150081) at 1/5000 dilution was used as the secondary antibody.



Flow Cytometry (Intracellular) - Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335)

Flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized mouse primary neuron cells labelling Neutrophil Elastase with ab310335 at 1/500 dilution (0.1 ug)/Right (Red) compared with a Rabbit monoclonal IgG (ab172730) / Left isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat Anti-Rabbit IgG (Alexa Fluor® 488, ab150081) at 1/5000 dilution was used as the secondary antibody.



Multiplex immunohistochemistry - Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335)

Multiplex immunohistochemistry analysis of formalin/PFA-fixed paraffin-embedded Mouse bone marrow tissue staining Neutrophil Elastase with ab310335 at a 1/500 (1.076 µg/ml) dilution, CD19 with ab245235 at a 1/1000 (0.444 µg/ml) dilution and CD3 with ab237721 at a 1/2000 (0.264 µg/ml) dilution followed by a ready to use secondary antibody Opal Polymer HRP Ms + Rb.

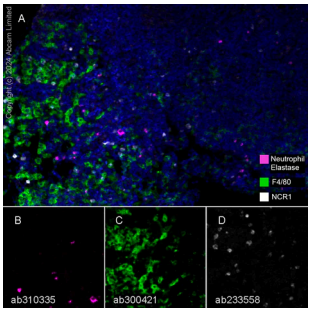
Panel A: merged staining of anti-Neutrophil Elastase (magenta; Opal™690), anti-CD19 (green; Opal™520) and anti-CD3 (gray; Opal™690) on mouse spleen.Panel B: anti-Neutrophil Elastase staining neutrophils in mouse spleen.
Panel C: anti-CD19 staining B lymphocytes in mouse spleen.
Panel D: anti-CD3 staining T lymphocytes in mouse spleen.
Nuclear DNA was labeled with DAPI (shown in blue).

The section was incubated in three rounds of staining: in the order of ab310335, ab245235 and ab237721 for 30 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system.

The immunostaining was performed on a Leica Biosystems BOND® RX instrument with an Opal™ 4-color kit. Image acquisition was performed with Leica SP8 confocal microscope.

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins

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Multiplex immunohistochemistry - Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335)

Neutrophil Elastase Multiplex immunohistochemistry staining of Mouse lymph node using rabbit Anti-Neutrophil Elastase antibody

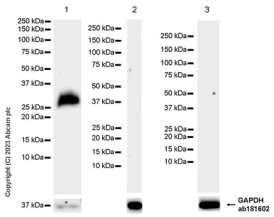
Multiplex immunohistochemistry analysis of formalin/PFA-fixed paraffin-embedded Mouse lymph node tissue staining Neutrophil Elastase with ab310335 at a 1/500 (1.076 µg/ml) dilution, F4/80 with ab300421 at a 1/5000 (0.106 µg/ml) dilution and NCR1 with ab233558 at a 1/500 (1.114 µg/ml) dilution followed by a ready to use secondary antibody Opal Polymer HRP Ms + Rb.

Panel A: merged staining of anti-Neutrophil Elastase (magenta; Opal™690), anti-CD19 (green; Opal™520) and anti-CD3 (gray; Opal™690) on mouse spleen.Panel B: anti-Neutrophil Elastase staining neutrophils in mouse spleen.
Panel C: anti-CD19 staining B lymphocytes in mouse spleen.
Panel D: anti-CD3 staining T lymphocytes in mouse spleen.
Nuclear DNA was labeled with DAPI (shown in blue).

The section was incubated in three rounds of staining: in the order of ab310335, ab245235 and ab237721 for 30 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system.

The immunostaining was performed on a Leica Biosystems BOND® RX instrument with an Opal™ 4-color kit. Image acquisition was performed with Leica SP8 confocal microscope.

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins



Western blot - Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335)

Blocking and diluting buffer and concentration: 5% NFDM/TBST Negative control: brain (PMID: 24309898), cerebellum (PMID: 24309898). In Western blot, anti-GAPDH antibody (ab181602) loading control staining at 1/200000 dilution. Exposure time: 48 seconds
All lanes:
Western blot - Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335) at 1/1000 dilution

Lane 1:
Human bone Marrow tissue lysate at 50 µg

Lane 2:
Human brain tissue lysate at 20 µg

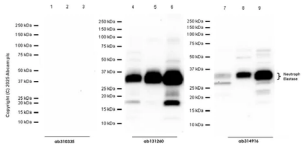
Lane 3:
Human cerebellum tissue lysate at 20 µg

Secondary

All lanes:
Goat Anti-Rabbit IgG (HRP) with minimal cross-reactivity with human IgG at 1/2000 dilution

Observed band size: 29 kDa

Exposure time: 48s



Western blot - Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335)

Neutrophil Elastase Western blot staining using rabbit Anti-Neutrophil Elastase antibody

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

ab310335 is not suitable for detecting human cell lines in western blot assay. ab131260 and ab314916 are alternatives for detecting human cell lines.

Neutrophil elastase undergoes proteolytic cleavage to generate fragments (PMID: 25857284).

Lanes 1 - 3:
Western blot - Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335) at 1/1000 dilution

Lanes 4 - 6:
Western blot - Anti-Neutrophil Elastase antibody [EPR7479] (ab131260) at 1/1000 dilution

Lanes 7 - 9:
Western blot - Anti-Neutrophil Elastase antibody [RM1077] (ab314916) at 1/1000 dilution

Lanes 1, 4 and 7:
HL-60 (human Acute Promyelocytic Leukemia promyeloblast) whole cell lysate at 20 µg

Lanes 2, 5 and 8:
THP-1 (human monocytic leukemia monocyte) whole cell lysate at 20 µg

Lanes 3, 6 and 9:
U937 (human histiocytic lymphoma monocyte) whole cell lysate at 20 µg

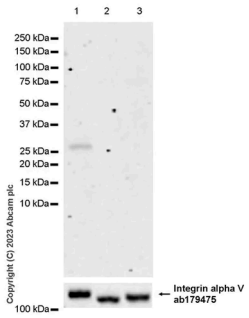
Secondary

All lanes:

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

Observed band size: 30 kDa, 26 kDa

Exposure time: 40s



Western blot - Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335)

Blocking and diluting buffer and concentration: 5% NFDM/TBST Negative controls: kidney (PMID: 24309898), cerebellum (PMID: 24309898). In Western blot, anti- Integrin alpha V antibody (ab179475) loading control staining at 1/5000 dilution. Exposure time: 180 seconds
All lanes:
Western blot - Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335) at 1/1000 dilution

Lane 1:
Rat bone marrow tissue lysate at 20 µg

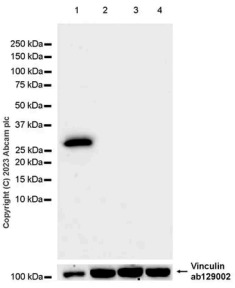
Lane 2:
Rat cerebellum tissue lysate at 20 µg

Lane 3:
Rat kidney tissue lysate at 20 µg

Secondary

All lanes:
Goat Anti-Rabbit IgG (HRP) with minimal cross-reactivity with human IgG at 1/2000 dilution

Exposure time: 180s



Western blot - Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335)

Neutrophil Elastase Western blot staining using rabbit Anti-Neutrophil Elastase antibody

Blocking and diluting buffer and concentration: 5% NFDM/TBST Negative controls: brain (PMID: 24309898), kidney (PMID: 24309898), cerebellum (PMID: 24309898). In Western blot, anti- Vinculin antibody (ab129002) loading control staining at 1/10000 dilution. Exposure time: 48 seconds
All lanes:
Western blot - Anti-Neutrophil Elastase antibody [EPR28386-66] (ab310335) at 1/1000 dilution

Lane 1:
Mouse bone marrow tissue lysate at 20 µg

Lane 2:
Mouse cerebellum tissue lysate at 20 µg

Lane 3:
Mouse kidney tissue lysate at 20 µg

Lane 4:
Mouse brain tissue lysate at 20 µg

Secondary

All lanes:
Goat Anti-Rabbit IgG (HRP) with minimal cross-reactivity with human IgG at 1/2000 dilution

Observed band size: 29 kDa

Exposure time: 48s

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