abcam

Product datasheet

Anti-NKG2A antibody [EPR23737-127] ab260035

Recombinant RabMAb

1 References 8 Images

Overview

Product name Anti-NKG2A antibody [EPR23737-127]

Description Rabbit monoclonal [EPR23737-127] to NKG2A

Host species Rabbit

Tested applications Suitable for: Flow Cyt (Intra), IHC-P, mIHC, WB

Species reactivity Reacts with: Human

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

Positive control WB: HEK-293 transfected with NKG2A (WT) expression vector containing a myc-His-tag, whole

cell lysate. IHC-P: Human tonsil, gastric cancer, NK cell lymphoma tissues and HEK-293

transfected with NKG2A expression vector containing a myc-His-tag®. Flow Cyt (intra): Human

peripheral blood mononuclear cells. mIHC: Human endometrium tissue.

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

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

Purity Protein A purified

Clonality Monoclonal

Clone number EPR23737-127

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Isotype IgG

Applications

The Abpromise guarantee Our <u>Abpromise guarantee</u> covers the use of ab260035 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)		1/500.
IHC-P		1/2000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
mIHC		1/2000.
WB		1/1000. Predicted molecular weight: 26 kDa.

Target

Function Plays a role as a receptor for the recognition of MHC class I HLA-E molecules by NK cells and

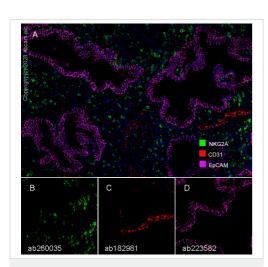
some cytotoxic T-cells.

Tissue specificity Natural killer cells.

Sequence similarities Contains 1 C-type lectin domain.

Cellular localization Membrane.

Images



Multiplex immunohistochemistry - Anti-NKG2A antibody [EPR23737-127] (ab260035)

Fluorescence multiplex immunohistochemical analysis of the human endometrium (Formalin/PFA-fixed paraffin-embedded sections).

Panel A: merged staining of anti-EpCAM (<u>ab223582</u>, magenta; Opal[™]690), anti-NKG2A (ab260035, green; Opal[™]520) and anti-CD31 (<u>ab182981</u>, red; Opal[™]570) on human endometrium. Panel B: anti-NKG2A stained on NK cells. Panel C: anti-CD31 stained on endothelial cells. Panel D: anti-EpCAM stained on glandular cells. Opal Polymer HRP Ms + Rb was used as a secondary antibody.

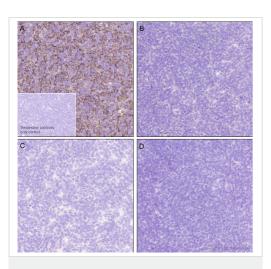
The section was incubated in three rounds of staining: in the order of $\underline{ab223582}$ at 1/500 dilution (1.008 µg/ml), ab260035 at 1/2000 dilution (0.262 µg/ml) and $\underline{ab182981}$ at 1/4000 dilution (0.137 µg/ml) for 30 mins at room temperature. Each round was followed by a separate fluorescent 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. DAPI (blue) was used as a nuclear counter stain.



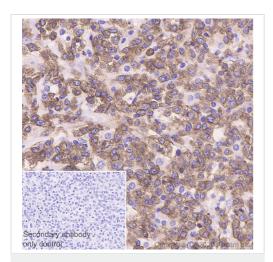
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-NKG2A antibody
[EPR23737-127] (ab260035)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of HEK-293 tissue labelling (A) - HEK-293 transfected with NKG2A expression vector containing a myc-Histag®, (B) - HEK-293 transfected with NKG2C expression vector containing a myc-His-tag®, (C) - HEK-293 transfected with NKG2E expression vector containing a myc-His-tag® whole cell pellets and (D) - HEK-293 transfected with an empty vector with ab260035 at 1/200 dilution. Heat mediated antigen retrieval was performed using Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins, Rabbit specific IHC polymer detection kit HRP/DAB (ab209101) was used as the secondary antibody (1/500). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.

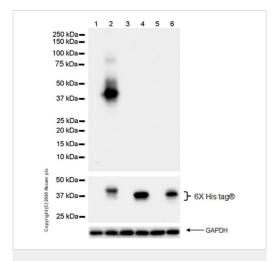
Positive staining on (A) HEK-293 transfected with NKG2A expression vector containing a myc-His-tag®, whole cell pellet. No staining on (B) HEK-293 transfected with NKG2C expression vector containing a myc-His-tag®, whole cell pellet and (C) HEK-293 transfected with NKG2E expression vector containing a myc-His-tag®, whole cell pellet. and (D) HEK-293 transfected with an empty vector.

The section was incubated with ab260035 for 30 mins at room temperature.

The immunostaining was performed on a Leica Biosystems BOND® RX instrument



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-NKG2A antibody
[EPR23737-127] (ab260035)



Western blot - Anti-NKG2A antibody [EPR23737-127] (ab260035)

Immunohistochemical analysis of paraffin-embedded Human NK cell lymphoma tissue labeling NKG2A with ab260035 at 1/2000 dilution followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101). Positive staining on human NK cell lymphoma (PMID: 11222501). The section was incubated with ab260035 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 Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).

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

All lanes : Anti-NKG2A antibody [EPR23737-127] (ab260035) at 1/5000 dilution

Lanes 1 & 3 & 5: HEK-293 (human embryonic kidney) transfected with an empty vector (vector control), containing a myc-His-tag®, whole cell lysate

Lane 2: HEK-293 transfected with NKG2A (WT) expression vector containing a myc-His-tag®, whole cell lysate

Lane 4: HEK-293 transfected with NKG2C (WT) expression vector containing a myc-His-tag®, whole cell lysate

Lane 6: HEK-293 transfected with NKG2E (WT) expression vector containing a myc-His-tag®, whole cell lysate

Lysates/proteins at 20 µg per lane.

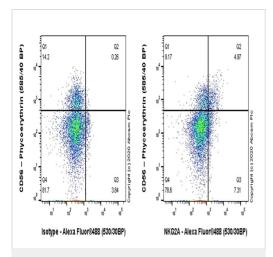
Secondary

All lanes : Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated (ab97051) at 1/50000 dilution

Predicted band size: 26 kDa Observed band size: 38 kDa

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

Exposure time: 3 seconds.

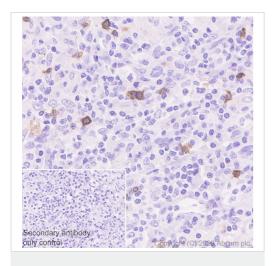


Flow Cytometry (Intracellular) - Anti-NKG2A antibody [EPR23737-127] (ab260035)

Intracellular flow cytometric analysis of 2% paraformaldehyde fixed, 0.1% Tween-20 permeabilized Human peripheral blood mononuclear cell (PBMC) cells labelling NKG2A with ab260035 at 1/500 dilution (0.1ug)/ Right compared with a Rabbit monoclonal lgG (ab172730) isotype control (Left).

A Goat anti rabbit lgG (Alexa Fluor[®] 488, **ab150077**) at 1/2000 dilution was used as the secondary antibody.

Cells were stained with anti-CD56 conjugated to PE, then fixed with 2% PFA followed by intracellularly stained with rabbit lgG (Left) or ab260035 (Right).

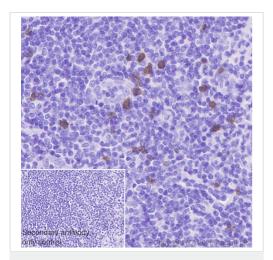


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-NKG2A antibody
[EPR23737-127] (ab260035)

Immunohistochemical analysis of paraffin-embedded Human gastric cancer tissue labeling NKG2A with ab260035 at 1/2000 dilution followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101). Positive staining on human gastric cancer (PMID: 11222501). The section was incubated with ab260035 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 Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).

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

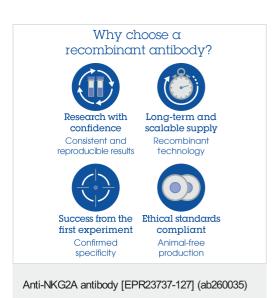


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-NKG2A antibody
[EPR23737-127] (ab260035)

Immunohistochemical analysis of paraffin-embedded Human tonsil tissue labeling NKG2A with ab260035 at 1/2000 dilution followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101). Positive staining on human tonsil (PMID: 11222501). The section was incubated with ab260035 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 Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).

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



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