abcam

Product datasheet

Anti-Granulin antibody [EPR15864] ab208777

KO VALIDATED Recombinant RabMAb

11 Images

Overview

Product name	Anti-Granulin antibody [EPR15864]
Description	Rabbit monoclonal [EPR15864] to Granulin
Host species	Rabbit
Tested applications	**Suitable for:** WB, IP, IHC-P
Species reactivity	**Reacts with:** Human
Immunogen	Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: HepG2, HAP1, A431 and 293T whole cell lysate; Human fetal kidney lysate; Human GRN full-length recombinant protein with GST-tag at N-terminal. IHC-P: Human liver, placenta, breast cancer and gastric cancer tissues. IP: HeLa 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 Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR15864

Isotype	IgG

Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab208777 in the following tested applications.

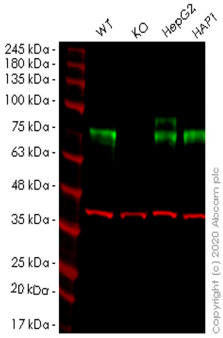
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB		1/2000. Detects a band of approximately 74 kDa (predicted molecular weight: 64 kDa).
IP		1/70.
IHC-P		1/1000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Target

Function	Granulins have possible cytokine-like activity. They may play a role in inflammation, wound repair, and tissue remodeling. Granulin-4 promotes proliferation of the epithelial cell line A431 in culture while granulin-3 acts as an antagonist to granulin-4, inhibiting the growth.
Tissue specificity	In myelogenous leukemic cell lines of promonocytic, promyelocytic, and proerythroid lineage, in fibroblasts, and very strongly in epithelial cell lines. Present in inflammatory cells and bone marrow. Highest levels in kidney.
Involvement in disease	Defects in GRN are the cause of ubiquitin-positive frontotemporal dementia (UP-FTD) [MIM:607485]; also known as tau-negative frontotemporal dementia linked to chromosome 17. Frontotemporal dementia (FTD) is the second most common cause of dementia in people under the age of 65 years. It is an autosomal dominant neurodegenerative disease.
Sequence similarities	Belongs to the granulin family.
Post-translational modifications	Granulins are disulfide bridged.
Cellular localization	Secreted.

Images



Western blot - Anti-Granulin antibody [EPR15864] (ab208777)

All lanes : Anti-Granulin antibody [EPR15864] (ab208777) at 1/1000 dilution

Lane 1 : Wild-type HEK293T cell lysate
Lane 2 : GRN knockout HEK293T cell lysate
Lane 3 : HepG2 cell lysate
Lane 4 : HAP1 cell lysate

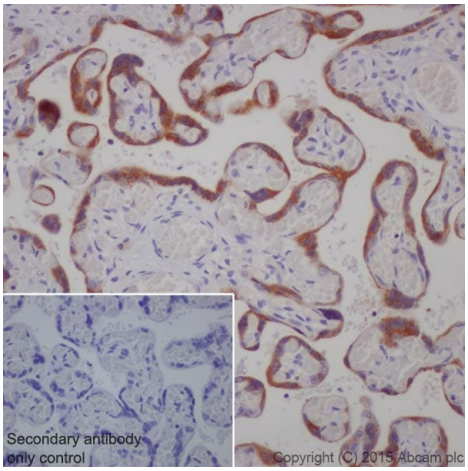
Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) at 1/10000 dilution

Predicted band size: 64 kDa
Observed band size: 74 kDa

Lanes 1-4: Merged signal (red and green). Green - ab208777 observed at 74 kDa. Red - loading control ab8245 observed at 36 kDa.

ab208777 Anti-Granulin antibody [EPR15864] was shown to specifically react with Granulin in wild-type HEK293T cells. Loss of signal was observed when knockout cell line ab266738 (knockout cell lysate ab257235) was used. Wild-type and Granulin knockout samples were subjected to SDS-PAGE. ab208777 and Anti-GAPDH antibody [6C5] - Loading Control (ab8245) were incubated at room temperature for 2.5 hours at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

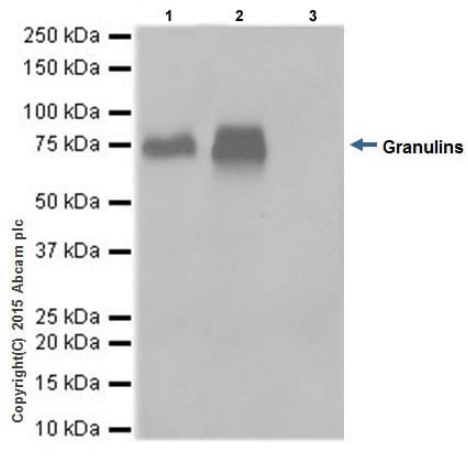


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Granulin antibody [EPR15864] (ab208777)

Immunohistochemical analysis of paraffin-embedded Human placenta tissue labeling Granulin with ab208777 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution. Cytoplasm staining on trophoblastic cells of Human placenta is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is ab97051 at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunoprecipitation - Anti-Granulin antibody [EPR15864] (ab208777)

Granulin was immunoprecipitated from 1mg of HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate with ab208777 at 1/70 dilution.

Western blot was performed from the immunoprecipitate using ab208777 at 1/1000 dilution.

VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/10000 dilution.

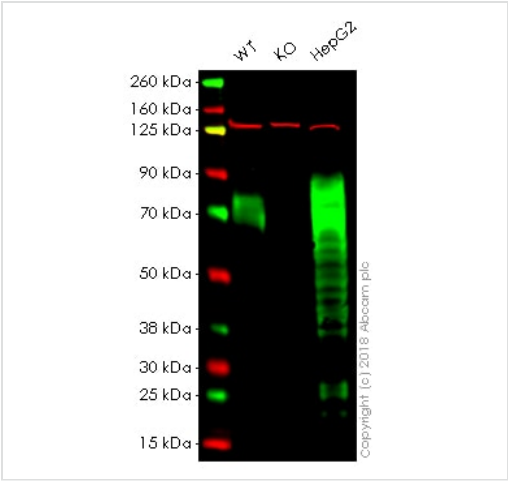
Lane 1: HeLa whole cell lysate, 10µg (Input).

Lane 2: ab208777 IP in HeLa whole cell lysate.

Lane 3: Rabbit IgG,monoclonal -Isotype Control (ab172730) instead of ab208777 in HeLa whole cell lysate.

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

Exposure time: 30 seconds.



Western blot - Anti-Granulin antibody [EPR15864] (ab208777)

All lanes : Anti-Granulin antibody [EPR15864] (ab208777) at 1/2000 dilution

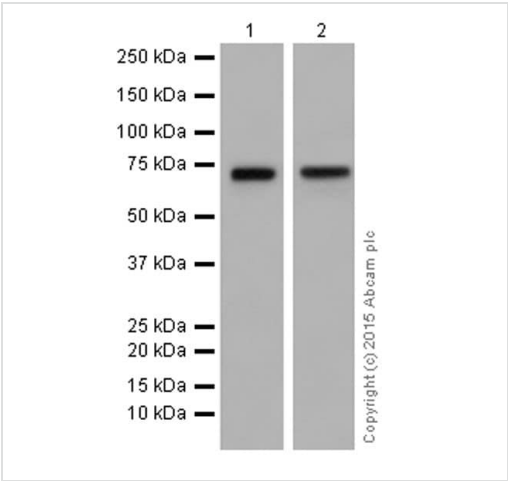
Lane 1 : Wild-type HAP1 whole cell lysate
Lane 2 : GRN (Granulin) knockout HAP1 whole cell lysate
Lane 3 : HepG2 whole cell lysate

Lysates/proteins at 20 µg per lane.

Predicted band size: 64 kDa

Lanes 1 - 3: Merged signal (red and green). Green - ab208777 observed at 64 kDa. Red - loading control, ab8245, observed at 37 kDa.

ab208777 was shown to specifically react with Granulin in wild-type HAP1 cells as signal was lost in GRN (Granulin) knockout cells. Wild-type and GRN (Granulin) knockout samples were subjected to SDS-PAGE. Ab208777 and ab8245 (Mouse anti GAPDH loading control) were incubated overnight at 4°C at 1/2000 dilution and 1/10000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed ab216773 and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed ab216776 secondary antibodies at 1/10000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-Granulin antibody [EPR15864] (ab208777)

Lane 1 : Anti-Granulin antibody [EPR15864] (ab208777) at 1/2000 dilution
Lane 2 : Anti-Granulin antibody [EPR15864] (ab208777) at 1/10000 dilution

Lane 1 : A431 (Human epidermoid carcinoma cell line) whole cell lysate
Lane 2 : 293T (Human epithelial cell line from embryonic kidney) whole cell lysate

Lysates/proteins at 10 µg per lane.

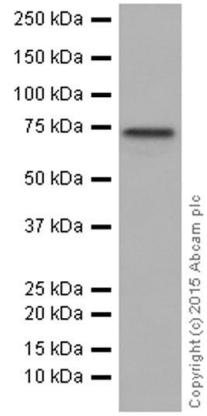
Secondary
All lanes : Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/1000 dilution

Predicted band size: 64 kDa

Observed band size: 74 kDa

Exposure time: 1 minute

Blocking/Dilution buffer: 5% NFDM/TBST.



Western blot - Anti-Granulin antibody [EPR15864] (ab208777)

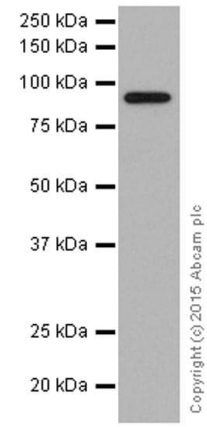
Anti-Granulin antibody [EPR15864] (ab208777) at 1/2000 dilution + Human fetal kidney lysate at 10 µg

Secondary
Anti-Rabbit IgG (HRP), specific to the non-reduced form of IgG at 1/1000 dilution

Predicted band size: 64 kDa
Observed band size: 74 kDa

Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDM/TBST.



Western blot - Anti-Granulin antibody [EPR15864] (ab208777)

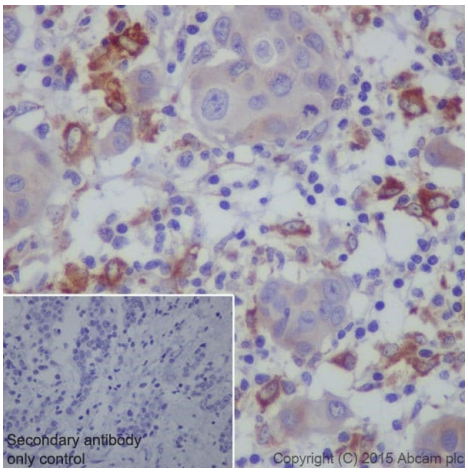
Anti-Granulin antibody [EPR15864] (ab208777) at 1/1000 dilution + Human GRN full-length recombinant protein with GST-tag at N-terminal at 0.01 µg

Secondary
Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/1000 dilution

Predicted band size: 64 kDa
Observed band size: 90 kDa

Exposure time: 15 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.

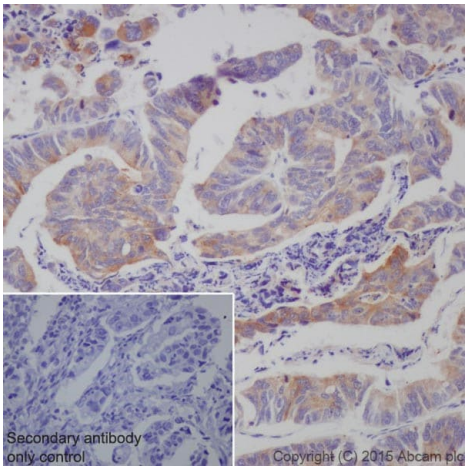


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Granulin antibody [EPR15864] (ab208777)

Immunohistochemical analysis of paraffin-embedded Human breast cancer tissue labeling Granulin with ab208777 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution. Cytoplasm staining on Human breast cancer is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is ab97051 at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

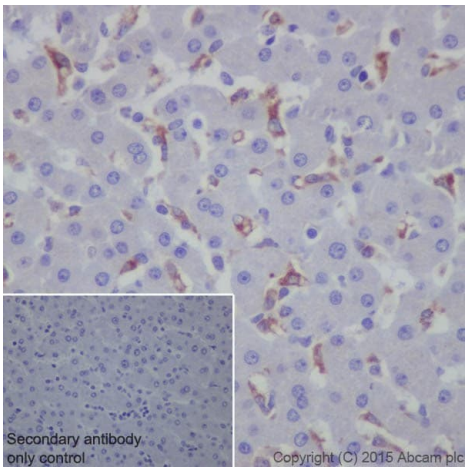


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Granulin antibody [EPR15864] (ab208777)

Immunohistochemical analysis of paraffin-embedded Human gastric cancer tissue labeling Granulin with ab208777 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution. Cytoplasm staining on Human gastric cancer is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is ab97051 at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

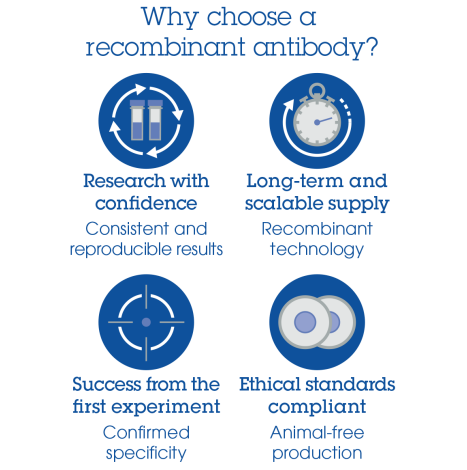


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Granulin antibody [EPR15864] (ab208777)

Immunohistochemical analysis of paraffin-embedded Human liver tissue labeling Granulin with ab208777 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution. Cytoplasm staining on Kupffer cells of Human liver is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is ab97051 at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Anti-Granulin antibody [EPR15864] (ab208777)

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