abcam

Product datasheet

Anti-Giantin antibody - Golgi Marker ab80864



Overview

Product name Anti-Giantin antibody - Golgi Marker

Description Rabbit polyclonal to Giantin - Golgi Marker

Host species Rabbit

Tested applications Suitable for: IHC-P, ICC/IF, WB

Species reactivity Reacts with: Human

Synthetic peptide corresponding to Human Giantin aa 1250-1350 conjugated to keyhole limpet **Immunogen**

haemocyanin.

(Peptide available as ab101412)

Positive control ICC/IF: MCF7 cells. IHC-P: Human kidney tissue.

General notes The Life Science industry has been in the grips of a reproducibility crisis for a number of years.

> Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets

your needs before purchasing.

If you have any questions, special requirements or concerns, please send us an inquiry and/or

contact our Support team ahead of purchase. Recommended alternatives for this product can be

found below, along with publications, customer reviews and Q&As

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 or -

80°C. Avoid freeze / thaw cycle.

Storage buffer pH: 7.40

Preservative: 0.02% Sodium azide

Constituent: PBS

Batches of this product that have a concentration < 1mg/ml may have BSA added as a stabilising

agent. If you would like information about the formulation of a specific lot, please contact our

scientific support team who will be happy to help.

Immunogen affinity purified **Purity**

1

Clonality

Polyclonal

Isotype

ΙgG

Applications

The Abpromise guarantee

Our $\underline{\textbf{Abpromise guarantee}}$ covers the use of ab80864 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
IHC-P	★★★★☆ (1)	Use a concentration of 0.1 - 1 µg/ml. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
ICC/IF	★★★★ <u>(1)</u>	Use a concentration of 1 µg/ml.
WB		1/1000. Predicted molecular weight: 376 kDa.

Target

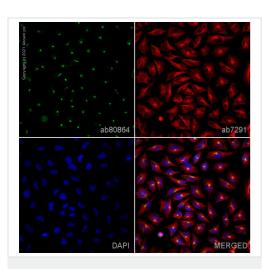
Function

May participate in forming intercisternal cross-bridges of the Golgi complex.

Cellular localization

Golgi apparatus membrane.

Images

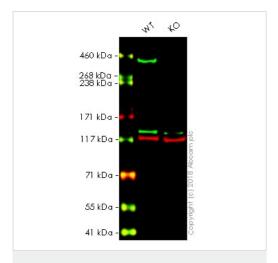


Immunocytochemistry/ Immunofluorescence - Anti-Giantin antibody - Golgi Marker (ab80864)

ab80864 staining Giantin in HeLa cells. The cells were fixed with 100% methanol (5 min), permeabilized with 0.1% PBS-Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1%PBS-Tween for 1h. The cells were then incubated overnight at 4°C with ab80864 at 1µg/ml and ab7291, Mouse monoclonal [DM1A] to alpha Tubulin - Loading Control. Cells were then incubated with ab150081, Goat polyclonal Secondary Antibody to Rabbit IgG - H&L (Alexa Fluor® 488), pre-adsorbed at 1/1000 dilution (shown in green) and ab150120, Goat polyclonal Secondary Antibody to Mouse IgG - H&L (Alexa Fluor® 594), pre-adsorbed at 1/1000 dilution (shown in pseudocolour red). Nuclear DNA was labelled with DAPI (shown in blue).

Also suitable in cells fixed with 4% paraformaldehyde (10 min).

Image was acquired with a high-content analyser (Operetta CLS, Perkin Elmer) and a maximum intensity projection of confocal sections is shown.



Western blot - Anti-Giantin antibody - Golgi Marker (ab80864)

All lanes : Anti-Giantin antibody - Golgi Marker (ab80864) at 1/1000 dilution

Lane 1: Wild-type HAP1 whole cell lysate

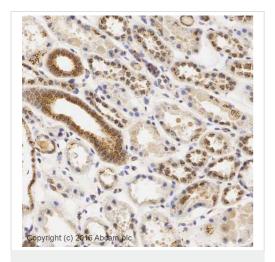
Lane 2: GOLGB1 knockout HAP1 whole cell lysate

Lysates/proteins at 20 µg per lane.

Predicted band size: 376 kDa

Lanes 1 - 2: Merged signal (red and green). Green - ab80864 observed at 377kDa. Red - loading control, <u>ab130007</u>, observed at 130kDa.

ab80864 was shown to recognize GOLGB1 in wild-type HAP1 cells as signal was lost at the expected MW in GOLGB1 knockout cells. Additional cross-reactive bands were observed in the wild-type and knockout cells. Wild-type and GOLGB1 knockout samples were subjected to SDS-PAGE. Ab80864 and ab130007 (Mouse anti-Vinculin loading control) were incubated overnight at 4°C at 1/1000 dilution and 1/20000 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 1/20000 dilution for 1 hour at room temperature before imaging.

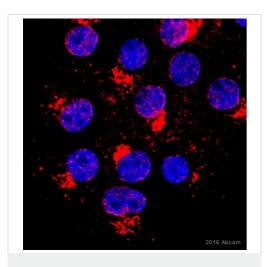


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Giantin antibody - Golgi Marker (ab80864)

IHC image of ab80864 staining in human kidney formalin fixed paraffin embedded tissue section*, performed on a Leica BondTM system using the standard protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with ab80864, 0.1µg/ml for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.

*Tissue obtained from the Human Research Tissue Bank, supported by the NIHR Cambridge Biomedical Research Centre



Immunocytochemistry/ Immunofluorescence - Anti-Giantin antibody - Golgi Marker (ab80864)

This image is courtesy of an anonymous Abreview

Ab80864 staining Giantin in WIF-B cells (hepatoma-derived hybrid cell line) by ICC/IF (Immunocytochemistry/Immunofluorescence). Cells were fixed with formaldehyde; permeabilized with 0.2% Triton X-100 and blocked with 1% Donkey serum in 0.1% PBST for 1 hour at 22°C. Samples were incubated at 1/25 dilution for 3 hours at 22°C. An Alexa Fluor[®] 594 Donkey anti rabbit was used as the secondary antibody at 1/200 dilution.

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