abcam

Product datasheet

Anti-N Cadherin antibody [EPR22397-264] - BSA and Azide free ab245827

KO VALIDATED Recombinant RabMAb

5 Images

Overview

Product name	Anti-N Cadherin antibody [EPR22397-264] - BSA and Azide free
Description	Rabbit monoclonal [EPR22397-264] to N Cadherin - BSA and Azide free
Host species	Rabbit
Tested applications	**Suitable for:** WB, Flow Cyt, IP **Unsuitable for:** ICC/IF or IHC-P
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: HeLa, PC-3, C6, A549, HEK-293T and HepG2 whole cell lysate. Human brain lysate. Mouse brain and heart lysate. Rat brain, heart and liver lysate. Flow Cyt: MCF7 cells. IP: N Cadherin IP in HeLa whole cell lysate.
General notes	ab245827 is the carrier-free version of **ab245117**. 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	EPR22397-264
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab245827 in the following tested applications.

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

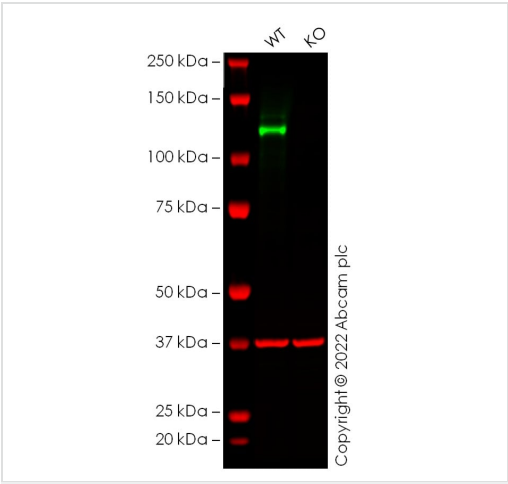
Application	Abreviews	Notes
WB		Use at an assay dependent concentration. Detects a band of approximately 130, 110 kDa (predicted molecular weight: 100 kDa).
Flow Cyt		Use at an assay dependent concentration.
IP		Use at an assay dependent concentration.

Application notes Is unsuitable for ICC/IF or IHC-P.

Target

Function	Cadherins are calcium dependent cell adhesion proteins. They preferentially interact with themselves in a homophilic manner in connecting cells; cadherins may thus contribute to the sorting of heterogeneous cell types. CDH2 may be involved in neuronal recognition mechanism. In hippocampal neurons, may regulate dendritic spine density.
Sequence similarities	Contains 5 cadherin domains.
Cellular localization	Cell membrane.

Images



Western blot - Anti-N Cadherin antibody [EPR22397-264] - BSA and Azide free (ab245827)

All lanes : Anti-N Cadherin antibody [EPR22397-264] (**ab245117**) at 1000 µg

Lane 1 : Wild-type HeLa cell lysate
Lane 2 : cdh2 knockout HeLa cell lysate

Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat anti-Rabbit IgG H&L 800CW and Goat anti-Mouse IgG H&L 680RD at 1/20000 dilution

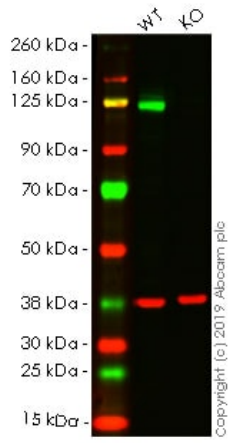
Performed under reducing conditions.

Predicted band size: 100 kDa
Observed band size: 125 kDa

This data was developed using the same antibody clone in a different buffer formulation (**ab245117**).

False colour image of Western blot: Anti-N Cadherin antibody [EPR22397-264] staining at 1/1000 dilution, shown in green; Mouse anti-GAPDH antibody [6C5] (**ab8245**) loading control staining at 1/20000 dilution, shown in red. In Western blot, **ab245117** was shown to bind specifically to N Cadherin. A band was observed at 125 kDa in wild-type HeLa cell lysates with no signal observed at this size in cdh2 knockout cell line **ab274934** (knockout cell lysate **ab274992**).

To generate this image, wild-type and cdh2 knockout HeLa cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween® 20 (TBS-T) before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit IgG H&L 800CW and Goat anti-Mouse IgG H&L 680RD at 1/20000 dilution.



Western blot - Anti-N Cadherin antibody [EPR22397-264] - BSA and Azide free (ab245827)

All lanes : Anti-N Cadherin antibody [EPR22397-264] (**ab245117**) at 1/100000 dilution

Lane 1 : Wild-type HEK-293T cell lysate
Lane 2 : CDH2 knockout HEK-293T cell lysate

Lysates/proteins at 20 µg per lane.

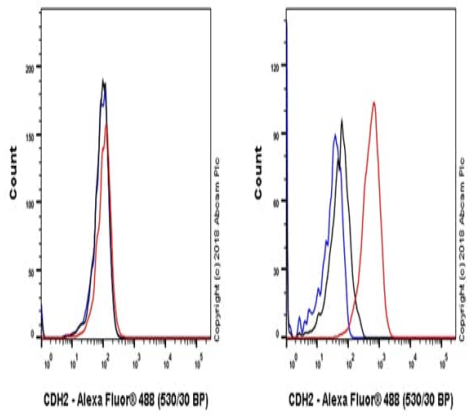
Performed under reducing conditions.

Predicted band size: 100 kDa
Observed band size: 130 kDa

This data was developed using the same antibody clone in a different buffer formulation (**ab245117**).

Lanes 1 - 2: Merged signal (red and green). Green - **ab245117** observed at 125 kDa. Red - loading control, **ab8245** observed at 37 kDa.

ab245117 was shown to react with N Cadherin in wild-type HEK-293T cells. Loss of signal was observed when knockout cell line **ab255377** (knockout cell lysate **ab263843**) was used. Wild-type and N Cadherin knockout samples were subjected to SDS-PAGE. **ab245117** and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) were incubated overnight at 4°C 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.



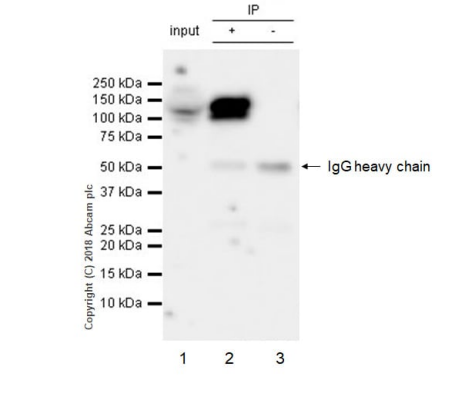
Flow Cytometry - Anti-N Cadherin antibody [EPR22397-264] - BSA and Azide free (ab245827)

Flow cytometric analysis of MCF7 (Human breast adenocarcinoma epithelial cell, Left) / HeLa (Human cervix adenocarcinoma epithelial cell, Right) cell lines labeling N Cadherin with **ab245117** at 1/500 (red) compared with a Rabbit monoclonal IgG (**ab172730**) (black) and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (blue). Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**), at 1/2000 dilution was used as the secondary antibody.

Negative control: MCF7 (PMID: 9177902).

Gated on viable cells.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab245117**).



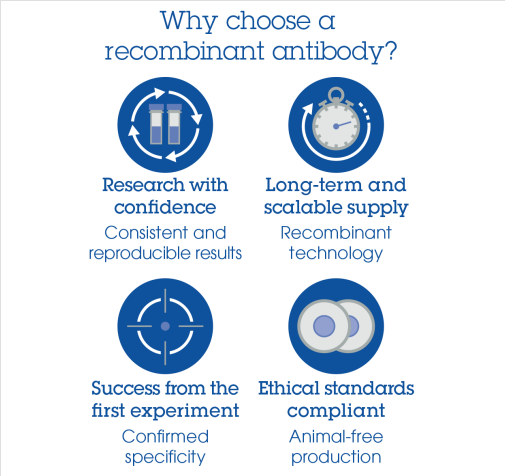
Immunoprecipitation - Anti-N Cadherin antibody [EPR22397-264] - BSA and Azide free (ab245827)

N Cadherin was immunoprecipitated from 0.35 mg HeLa (human cervix adenocarcinoma epithelial cell line) whole cell lysate with **ab245117** at 1/30 dilution. Western blot was performed from the immunoprecipitate using **ab245117** at 1/500 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**), was used for detection at 1/1000 dilution.

Lane 1: HeLa whole cell lysate 10 µg (Input).
Lane 2: **ab245117** IP in HeLa whole cell lysate.
Lane 3: Rabbit monoclonal IgG (**ab172730**) instead of **ab245117** in HeLa whole cell lysate.
Blocking/Dilution buffer: 5% NFDM/TBST.
Exposure time: 3 seconds.

The molecular weight is consistent with literature (PMID: 8230319).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab245117**).



Anti-N Cadherin antibody [EPR22397-264] - BSA and Azide free (ab245827)

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