abcam

Product datasheet

Anti-NDRG1 antibody [EPR5593] - Low endotoxin, Azide free ab216457



リコンピナント

RabMAb

画像数 13

製品の概要

製品名 Anti-NDRG1 antibody [EPR5593] - Low endotoxin, Azide free

製品の詳細 Rabbit monoclonal [EPR5593] to NDRG1 - Low endotoxin, Azide free

由来種 Rabbit

特異性 PBS only lot tested.

アプリケーション 適用あり: IP, IHC-P, WB, ICC/IF, Flow Cyt (Intra)

種交差性 交差種: Mouse, Rat, Human

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

ポジティブ・コントロール IHC-P: Human colon tissue, human liver carcinoma tissue, Mouse and rat colon tissue. ICC/IF:

Jurkat (Human T cell leukemia T lymphocyte) cells. IP: HeLa. WB: Wild-type HEK-293 whole cell lysate. Jurkat, HeLa, Caco-2 and LnCap whole cell lysate. Mouse and rat brain lysate. Flow

cyto(intra): HeLa (Human cervix adenocarcinoma epithelial cell)

特記事項 ab216457 is the carrier-free version of ab124689.

Our <u>carrier-free</u> 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**.

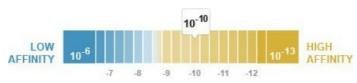
Our <u>Low endotoxin, azide-free formats</u> have low endotoxin level (≤ 1 EU/ml, determined by the LAL assay) and are free from azide, to achieve consistent experimental results in functional assays.

製品の特性

製品の状態 Liquid

保存方法 Shipped at 4°C. Store at +4°C. Do Not Freeze.

解離定数(K_D 値) $K_D = 1.33 \times 10^{-10} M$



Learn more about KD

バッファー pH: 7.2

Constituent: PBS

キャリア・フリー はい

精製度 Protein A purified

ポリ/モノ モノクローナル

クローン名 EPR5593

アイソタイプ lgG

アプリケーション

The Abpromise guarantee <u>Abpromise保証は、</u>次のテスト済みアプリケーションにおけるab216457の使用に適用されます アプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
IP		Use at an assay dependent concentration.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
WB		Use at an assay dependent concentration. Detects a band of approximately 48 kDa (predicted molecular weight: 43 kDa).
ICC/IF		Use at an assay dependent concentration.
Flow Cyt (Intra)		Use at an assay dependent concentration.

機能

組織特異性

関連疾患

配列類似性 細胞内局在

能 May have a growth inhibitory role.

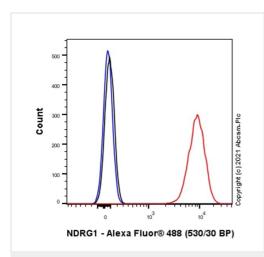
Ubiquitous; expressed most prominently in placental membranes and prostate, kidney, small intestine, and ovary tissues. Reduced expression in adenocarcinomas compared to normal tissues. In colon, prostate and placental membranes, the cells that border the lumen show the highest expression.

Defects in NDRG1 are the cause of Charcot-Marie-Tooth disease type 4D (CMT4D) [MIM:601455]; also known as hereditary motor and sensory neuropathy Lom type (HMSNL). CMT4D is a recessive form of Charcot-Marie-Tooth disease, the most common inherited disorder of the peripheral nervous system. Charcot-Marie-Tooth disease is classified in two main groups on the basis of electrophysiologic properties and histopathology: primary peripheral demyelinating neuropathy and primary peripheral axonal neuropathy. Demyelinating CMT neuropathies are characterized by severely reduced nerve conduction velocities (less than 38 m/sec), segmental demyelination and remyelination with onion bulb formations on nerve biopsy, slowly progressive distal muscle atrophy and weakness, absent deep tendon reflexes, and hollow feet. By convention, autosomal recessive forms of demyelinating Charcot-Marie-Tooth disease are designated CMT4.

Belongs to the NDRG family.

Cytoplasm. Nucleus. Cell membrane. Whereas in prostate epithelium and placental chorion it is located in both the cytoplasm and the nucleus, nuclear staining is not observed in colon epithelium cells. Instead its localization changes from the cytoplasm to the plasma membrane during differentiation of colon carcinoma cell lines in vitro.

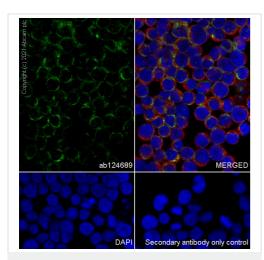
画像



Flow Cytometry (Intracellular) - Anti-NDRG1 antibody [EPR5593] - Low endotoxin, Azide free (ab216457)

This data was developed using <u>ab124689</u>, the same antibody clone in a different buffer formulation.

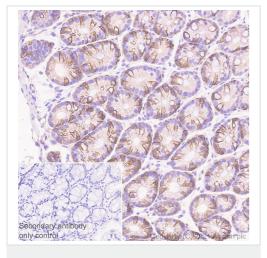
Intracellular Flow Cytometry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling NDRG1 with purified ab124689 at 1/20 dilution (5 ug/ml) (Red). Cells were fixed with 4% Paraformaldehyde and permeabilised with 90% Methanol. A Goat anti rabbit lgG (Alexa Fluor® 488, ab150081) (1/2000 dilution) was used as the secondary antibody. Rabbit monoclonal lgG (Black) was used as a isotype control. Cell without incubation with primary antibody and secondary antibody (Blue) were used as unlabeled control.



Immunocytochemistry/ Immunofluorescence - Anti-NDRG1 antibody [EPR5593] - Low endotoxin, Azide free (ab216457)

This data was developed using <u>ab124689</u>, the same antibody clone in a different buffer formulation.

Immunocytochemistry/ Immunofluorescence analysis of Jurkat (Human T cell leukemia T lymphocyte) cells labeling NDRG1 using **ab124689**. The cells were fixed with 100% Methanol then permeabilized with 0.1% Triton X-100. The cells were then incubated with **ab124689** at 1:50 dilution followed by a further incubation with a Goat anti rabbit lgG (Alexa Fluor® 488, **ab150077**) at 2 μg/ml (shown in green). Nuclear DNA was labelled in blue with DAPI. Cells were counterstained using **ab195889** Antialpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) at 1:200 dilution (shown in red). Secondary antibody only control: PBS instead of the primary antibody.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-NDRG1 antibody

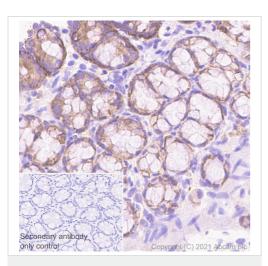
[EPR5593] - Low endotoxin, Azide free (ab216457)

This data was developed using <u>ab124689</u>, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of Paraffin-embedded sections mouse colon tissue labelling NDRG1 with ab124689 at 1/1000 dilution, followed by a ready to use secondary Rabbit specific IHC polymer detection kit HRP/DAB (ab209101). Staining on mouse colon tissue is observed. Counter stained with Haematoxylin. Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).

Heat mediated antigen retrieval using Bond™ Epitope Retrieval Solution 2 (pH 9.0).

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



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-NDRG1 antibody

[EPR5593] - Low endotoxin, Azide free (ab216457)

Secondary antibody

Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-NDRG1 antibody

[EPR5593] - Low endotoxin, Azide free (ab216457)

This data was developed using <u>ab124689</u>, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of Paraffin-embedded sections rat colon tissue labelling NDRG1 with <u>ab124689</u> at 1/1000 dilution, followed by a ready to use secondary Rabbit specific IHC polymer detection kit HRP/DAB (<u>ab209101</u>). Staining on rat colon tissue is observed. Counter stained with Haematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).

Heat mediated antigen retrieval using Bond™ Epitope Retrieval Solution 2 (pH 9.0).

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

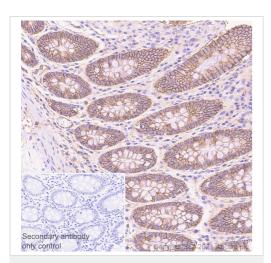
This data was developed using <u>ab124689</u>, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of Paraffin-embedded sections human liver carcinoma tissue labelling NDRG1 with **ab124689** at 1/1000 dilution, followed by a ready to use secondary Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**). Staining on human liver carcinoma tissue is observed. Counter stained with Haematoxylin.

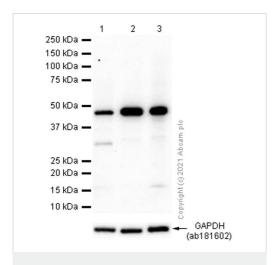
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).

Heat mediated antigen retrieval using Bond™ Epitope Retrieval Solution 2 (pH 9.0).

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



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-NDRG1 antibody
[EPR5593] - Low endotoxin, Azide free (ab216457)



Western blot - Anti-NDRG1 antibody [EPR5593] - Low endotoxin, Azide free (ab216457)

This data was developed using <u>ab124689</u>, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of Paraffin-embedded sections human colon tissue labelling NDRG1 with <u>ab124689</u> at 1/1000 dilution, followed by a ready to use secondary Rabbit specific IHC polymer detection kit HRP/DAB (<u>ab209101</u>). Staining on human colon tissue is observed. Counter stained with Haematoxylin. Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is ready to use Rabbit specific IHC polymer detection kit HRP/DAB (<u>ab209101</u>).

Heat mediated antigen retrieval using Bond™ Epitope Retrieval Solution 2 (pH 9.0).

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

All lanes : Anti-NDRG1 antibody [EPR5593] (ab124689) at 1/10000 dilution

Lane 1 : HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate

Lane 2 : Mouse brain lysate

Lane 3 : Rat brain lysate

Lysates/proteins at 20 µg per lane.

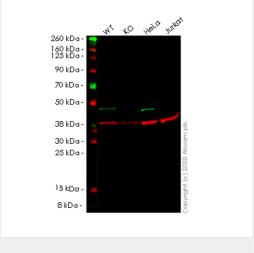
Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

Predicted band size: 43 kDa **Observed band size:** 48 kDa

This data was developed using <u>ab124689</u>, the same antibody clone in a different buffer formulation.

Blocking and diluting buffer and concentration: 5% NFDM/TBST. **ab181602** was used as GAPDH loading control.



Western blot - Anti-NDRG1 antibody [EPR5593] - Low endotoxin, Azide free (ab216457)

All lanes : Anti-NDRG1 antibody [EPR5593] (<u>ab124689</u>) at 1/1000 dilution

Lane 1: Wild-type HEK-293T (Human epithelial cell line from embryonic kidney transformed with large T antigen) whole cell lysate

Lane 2: NDRG1 knockout HEK-293T (Human epithelial cell line from embryonic kidney transformed with large T antigen) whole cell lysate

Lane 3 : HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate

Lane 4: Jurkat (Human T cell leukemia cell line from peripheral blood) whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

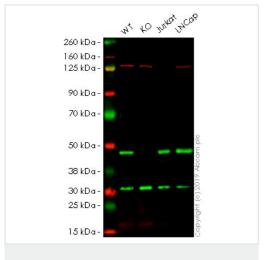
All lanes : Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (ab216773) at 1/10000 dilution

Predicted band size: 43 kDa
Observed band size: 43 kDa

This data was developed using <u>ab124689</u>, the same antibody clone in a different buffer formulation.

Lanes 1-4: Merged signal (red and green). Green - <u>ab124689</u> observed at 43 kDa. Red - loading control <u>ab8245</u> observed at 36 kDa.

<u>ab124689</u> Anti-NDRG1 antibody [EPR5593] was shown to specifically react with NDRG1 in wild-type HEK-293T cells. Loss of signal was observed when knockout cell line <u>ab267301</u> (knockout cell lysate <u>ab257551</u>) was used. Wild-type and NDRG1 knockout samples were subjected to SDS-PAGE. <u>ab124689</u> and Anti-GAPDH antibody [6C5] - Loading Control (<u>ab8245</u>) were incubated overnight at 4°C at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (<u>ab216773</u>) and Goat anti-Mouse lgG H&L (IRDye® 680RD) preadsorbed (<u>ab216776</u>) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-NDRG1 antibody [EPR5593] - Low endotoxin, Azide free (ab216457)

All lanes : Anti-NDRG1 antibody [EPR5593] (ab124689) at 1/10000 dilution

Lane 1 : Wild-type HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate

Lane 2: NDRG1 knockout HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate

Lane 3 : Jurkat (Human T cell leukemia cell line from peripheral blood) whole cell lysate

Lane 4: LNCaP (Human prostate cancer cell line) whole cell lysate

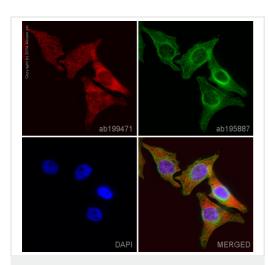
Lysates/proteins at 20 µg per lane.

Predicted band size: 43 kDa

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab124689**).

Lanes 1 - 4: Merged signal (red and green). Green - <u>ab124689</u> observed at 43 kDa. Red - loading control, <u>ab130007</u>, observed at 130 kDa.

ab124689 was shown to recognize in wild-type HEK-293 cells as signal was lost at the expected MW in NDRG1 knockout cells. Additional cross-reactive bands were observed in the wild-type and knockout cells. Wild-type and NDRG1 knockout samples were subjected to SDS-PAGE. Ab124689 and ab130007 (Mouse anti-Vinculin loading control) were incubated overnight at 4°C at 1/10000 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.



Immunocytochemistry/ Immunofluorescence - Anti-NDRG1 antibody [EPR5593] - Low endotoxin, Azide free (ab216457)

ab199233 ab195889

Immunocytochemistry/ Immunofluorescence - Anti-NDRG1 antibody [EPR5593] - Low endotoxin, Azide free (ab216457)

Clone EPR5593 (ab216457) has been successfully conjugated by Abcam. This image was generated using Anti-NDRG1 antibody [EPR5593] (Alexa Fluor® 647). Please refer to **ab199471** for protocol details.

<u>ab199471</u> staining NDRG1 in HeLa cells. The cells were fixed with 4% formaldehyde (10 min), permeabilized with 0.1% 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 ab at a 1/100 dilution (shown in red) and <u>ab195887</u>, Mouse monoclonal to alpha Tubulin (Alexa Fluor[®] 488), at a 1/250 dilution (shown in green). Nuclear DNA was labelled with DAPI (shown in blue).

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

Clone EPR5593 (ab216457) has been successfully conjugated by Abcam. This image was generated using Anti-NDRG1 antibody [EPR5593] (Alexa Fluor® 488). Please refer to ab199233 for protocol details.

ab199233 staining NDRG1 in HeLa cells. The cells were fixed with 4% formaldehyde (10 min), permeabilized with 0.1% 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 **ab199233** at 1/500 dilution (shown in green) and **ab195889**, Mouse monoclonal to alpha Tubulin (Alexa Fluor[®] 594), at 1/250 dilution (shown in red). Nuclear DNA was labelled with DAPI (shown in blue).

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).



Immunoprecipitation - Anti-NDRG1 antibody
[EPR5593] - Low endotoxin, Azide free (ab216457)

Lane 1 (input): HeLa(Human cervix adenocarcinoma epithelial cell) whole cell lysate 10µg

Lane 2 (+): ab124689 & HeLa whole cell lysate

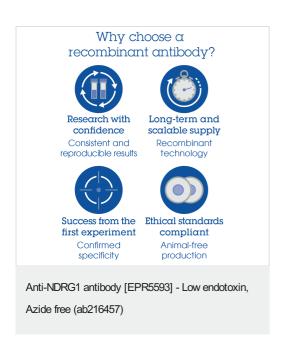
Lane 3 (-): Rabbit monoclonal IgG (ab172730) instead of

ab124689 in HeLa whole cell lysate

Ab124689 (Purified) at 1:500 dilution (1.86 μg/ml) immunoprecipitating NDRG1 in HeLa whole cell lysate. For western blotting, VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/1,000 dilution.

Blocking and diluting buffer: 5% NFDM /TBST.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab124689).



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