abcam

Product datasheet

Anti-Nrf2 antibody [EP1808Y] - ChIP Grade ab62352

重组 RabMAb

★★★★ 26 Abreviews 304 References 13 图像

概述

产品名称 Anti-Nrf2抗体[EP1808Y] - ChIP Grade

宿主 Rabbit

特异性 The expression of Nrf2 is stimulated by oxidative stress, electrophiles and chemical

activators (PMID: 25761198, PMID: 27638861 and PMID: 28587109). Nrf2 antibody (ab62352) detects no signal in most untreated samples in WB. Stimuli treated samples are recommended. We do not recommend using this product in western blot with tissue

lysates, however some customers have used this antibody successfully using

concentrated samples (see submitted abreviews).

经测试应用 适用于: Flow Cyt, ChIP, ICC/IF, WB, IP, IHC-P

种属反应性 与反应: Human

免疫原 Synthetic peptide within Human Nrf2 aa 550 to the C-terminus (C terminal). The exact sequence

is proprietary.

Database link: Q16236

(Peptide available as ab167152)

阳性对照 WB: MG-132 treated HeLa whole cell lysate, Saos-2, THP-1, MG-132 treated HepG2 whole cell

lysate, MG-132 treated HCT-116 whole cell lysate. IHC-P: Human pancreatic carcinoma and kidney cancer tissues. ICC/IF: HepG2 and HeLa cells. Flow Cyt: HeLa cells. IP: SAOS-2 whole

cell lysate. ChIP: Chromatin prepared from HepG2 cells.

常规说明 PLEASE NOTE:

Nrf2 antibody (ab62352) detects no signal in most untreated samples for WB. Stimuli treated samples are recommended. Nrf2 expression is stimulated by oxidative stress, electrophiles and chemical activators (PMID: 25761198, PMID: 27638861 and PMID:

28587109)

Mouse is not recommended for IHC (weak staining).

Rat: We have preliminary internal testing data to indicate this antibody may not react with these species. Please contact us for more information.

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

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- 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.

性能

形式 Liquid

存放说明 Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C.

Avoid freeze / thaw cycle.

存储溶液 pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: PBS, 40% Glycerol, 0.05% BSA

纯**度** Protein A purified

同种型 lgG

应用

The Abpromise guarantee Abpromise™承诺保证使用ab62352于以下的经测试应用

"应用说明"部分下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
Flow Cyt		1/40.
ChIP	★★★★ (1)	Use 2 µg for 25 µg of chromatin.
ICC/IF	★★★★★ (4)	1/100 - 1/500.
WB	★★★★☆ (19)	1/200 - 1/1000. Predicted molecular weight: 68 kDa.Can be blocked with Nrf2 peptide (ab167152).
IP		1/20.
IHC-P	★★★★★ (1)	1/100 - 1/250. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

靶标

功能

Transcription activator that binds to antioxidant response (ARE) elements in the promoter regions of target genes. Important for the coordinated up-regulation of genes in response to oxidative stress. May be involved in the transcriptional activation of genes of the beta-globin cluster by mediating enhancer activity of hypersensitive site 2 of the beta-globin locus control region.

组织特异性 Widely expressed. Highest expression in adult muscle, kidney, lung, liver and in fetal muscle.

序列相似性 Belongs to the bZIP family. CNC subfamily.

Contains 1 bZIP domain.

结构域 Acidic activation domain in the N-terminus, and DNA binding domain in the C-terminus.

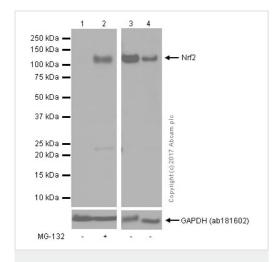
翻译后修饰 Phosphorylation of Ser-40 by PKC in response to oxidative stress dissociates NFE2L2 from its

cytoplasmic inhibitor KEAP1, promoting its translocation into the nucleus.

细胞定位 Cytoplasm > cytosol. Nucleus. Cytosolic under unstressed conditions, translocates into the

nucleus upon induction by electrophilic agents.

图片



Western blot - Anti-Nrf2 antibody [EP1808Y] - ChIP Grade (ab62352)

All lanes : Anti-Nrf2 antibody [EP1808Y] - ChIP Grade (ab62352) at 1/1000 dilution (purified)

Lane 1: HeLa whole cell lysate

Lane 2: HeLa whole cell lysate treated with MG-132 2 µM for 18

hours

Lane 3 : Saos-2 whole cell lysate

Lane 4 : THP-1 whole cell lysate

Lysates/proteins at 15 µg per lane.

Secondary

All lanes: Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/20000

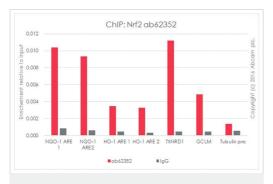
dilution

Predicted band size: 68 kDa **Observed band size:** 100 kDa

Blocking and dilution buffer: 5% NFDM/TBST.

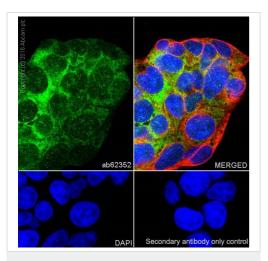
Exposure:

Lanes 1 and 2: 15 seconds. Lanes 3 and 4: 3 minutes.



ChIP - Anti-Nrf2 antibody [EP1808Y] - ChIP Grade (ab62352)

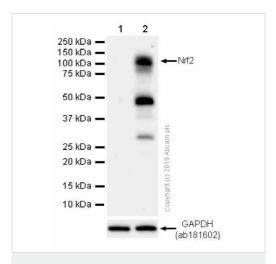
Chromatin was prepared from HepG2 cells according to the Abcam X-ChIP protocol. Cells were fixed with 1% formaldehyde for 10 minutes. The ChIP was performed with 25µg of chromatin, 2µg of ab62352 (blue), and 20µl of A/G sepharose bead slurry (10µl of sepharose A beads + 10µl of sepharose G beads). 2µg of rabbit normal lgG was added to the beads control (yellow). The immunoprecipitated DNA was quantified by real time PCR (Sybr green approach).



Immunocytochemistry/ Immunofluorescence - Anti-Nrf2 antibody [EP1808Y] - ChIP Grade (ab62352)

Immunocytochemistry/Immunofluorescence analysis of HepG2 cells labelling Nrf2 with purified ab62352 at 1/500. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. ab150077, an Alexa Fluor[®] 488-conjugated goat anti-rabbit lgG (1/500) was used as the secondary antibody. The cells were costained with ab195889, an Alexa Fluor[®] 594-conjugated mouse anti-alpha tubulin antibody (1/200). Nuclei counterstained with DAPI (blue).

Secondary antibody only control: PBS was used instead of the primary antibody as the negative control.



Western blot - Anti-Nrf2 antibody [EP1808Y] - ChIP Grade (ab62352)

All lanes : Anti-Nrf2 antibody [EP1808Y] - ChIP Grade (ab62352) at 1/200 dilution (Purified)

Lane 1 : HCT 116 (Human colorectal carcinoma epithelial cell) whole cell lysates with 5% NFDM/TBST

Lane 2: HCT 116 (Human colorectal carcinoma epithelial cell) treated with 25uM MG-132 for 4 hours whole cell lysates with 5% NFDM/TBST

Lysates/proteins at 20 µg per lane.

Secondary

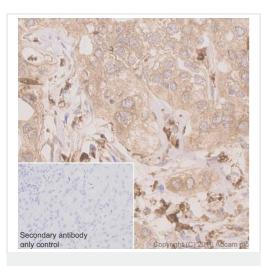
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

Predicted band size: 68 kDa

Observed band size: 100 kDa

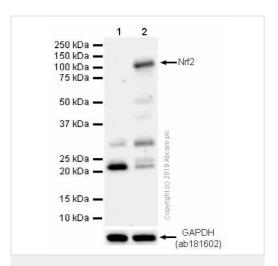
Exposure time: 60 seconds

The expression of Nrf2 is stimulated by oxidative stress, electrophiles and chemical activators (PMID: 25761198, PMID: 27638861 and PMID: 28587109). ab62352 detects no signal in most of the untreated samples in WB. Stimuli treated samples are recommended.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Nrf2 antibody [EP1808Y] - ChIP Grade (ab62352)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human pancreatic carcinoma tissue labelling Nrf2 with purified ab62352 at a dilution of 1/250. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9 (ab93684). Goat Anti-Rabbit IgG H&L (HRP) (ab97051) was used as the secondary antibody (1/500). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.



Western blot - Anti-Nrf2 antibody [EP1808Y] - ChIP Grade (ab62352)

All lanes : Anti-Nrf2 antibody [EP1808Y] - ChIP Grade (ab62352) at 1/200 dilution (Purified)

Lane 1 : HepG2 (Human hepatocellular carcinoma epithelial cell) whole cell lysates with 5% NFDM/TBST

Lane 2 : HepG2 (Human hepatocellular carcinoma epithelial cell) treated with 2uM MG-132 for 18 hours whole cell lysates with 5% NFDM/TBST

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/20000 dilution

Predicted band size: 68 kDa

Observed band size: 100 kDa

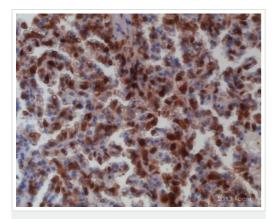
Exposure time: 180 seconds

The expression of Nrf2 is stimulated by oxidative stress, electrophiles and chemical activators (PMID: 25761198, PMID: 27638861 and PMID: 28587109). ab62352 detects no signal in most of the untreated samples in WB. Stimuli treated samples are recommended.

250 kDa —
150 kDa —
150 kDa —
75 kDa —
37 kDa —
25 kDa —
20 kDa —
16 kDa —
16 kDa —
16 kDa —
17 kDa —
18 kDa —
18 kDa —
19 kDa —
10 kDa —

Western blot - Anti-Nrf2 antibody [EP1808Y] - ChIP Grade (ab62352)

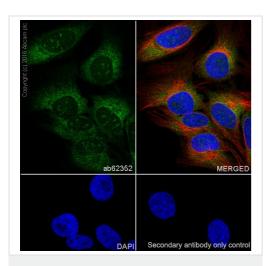
Different batches of ab62352 were tested on Saos-2 (Human osteosarcoma epithelial) lysate at 1.0 μ g/ml. 15 μ g of lysate was loaded in each lane. Bands observed at 100 kDa.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Nrf2 antibody [EP1808Y] - ChIP Grade (ab62352)

This image is courtesy of an abreview submitted by Rudolf Jung.

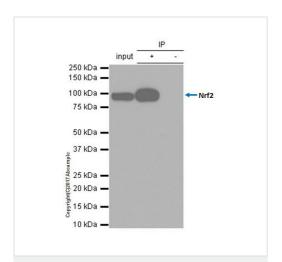
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human kidney cancer tissue sections labeling Nrf2 with ab62352 at 1/100 dilution. The tissue was fixed with paraformaldehyde and a heat mediated antigen retrival step was performed with TRIS-EDTA Buffer pH 9.0. Staining with ab62352 at 1/100 was carried out in a dilution buffer with blocking for 30 minutes at 20°C. A undiluted goat anti-rabbit HRP conjugated secondary antibody was used.



Immunocytochemistry/ Immunofluorescence - Anti-Nrf2 antibody [EP1808Y] - ChIP Grade (ab62352)

Immunocytochemistry/Immunofluorescence analysis of HeLa cells labelling Nrf2 with purified ab62352 at 1/500. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077) secondary antibody (1/1000) was used as the secondary antibody. Cells were counterstained with ab195889, anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594). DAPI was used to stain the nuclei blue.

Secondary antibody only control: PBS was used instead of the primary antibody as the negative control.



Immunoprecipitation - Anti-Nrf2 antibody [EP1808Y] - ChIP Grade (ab62352)

ab62352 (purified) at 1/30 immunoprecipitating Nrf2 in SAOS-2 whole cell lysate.

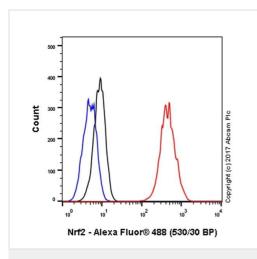
Lane 1 (input): SAOS-2 whole cell lysate (10µg)

Lane 2 (+): ab62352 + SAOS-2 whole cell lysate.

Lane 3 (-): Rabbit monoclonal lgG (ab172730) instead of ab62352 in SAOS-2 whole cell lysate.

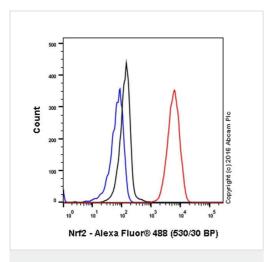
For western blotting, ab62352 was used at a dilution of 1/500 and VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/1000 dilution.

Blocking and dilution buffer: 5% NFDM/TBST.



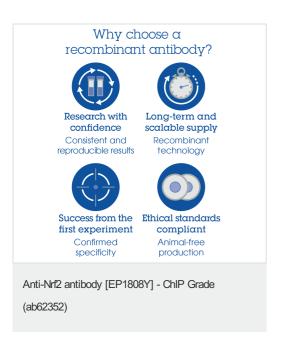
Flow Cytometry (Intracellular) - Anti-Nrf2 antibody [EP1808Y] - ChIP Grade (ab62352)

Flow Cytometry analysis of HeLa cells labelling Nrf2 with purified ab62352 at a dilution of 1/60 (red). Cells were fixed with 4% paraformaldehyde. Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077) secondary antibody (1/2000) was used as the secondary antibody. Black - Isotype control, rabbit monoclonal IgG (ab172730). Blue - Unlabelled control, cells without incubation with primary and secondary antibodies.



Flow Cytometry (Intracellular) - Anti-Nrf2 antibody [EP1808Y] - ChIP Grade (ab62352)

Flow Cytometry analysis of HeLa (human cervix adenocarcinoma) cells labelling Nrf2 with ab62352 at 1/40 (red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. An Alexa Fluor[®] 488-conjugated goat anti-rabbit lgG (1/2000) was used as the secondary antibody. Black - Isotype control, rabbit monoclonal lgG. Blue - Unlabelled control, cells without incubation with primary and secondary antibodies.



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