abcam

Product datasheet

Anti-Nrf2 (phospho S40) antibody [EP1809Y] ab76026



重组 RabMAb

★★★★ 18 Abreviews 180 References 17 图像

概述

产品名称 Anti-Nrf2 (phospho S40)抗体[EP1809Y]

描述 兔单克隆抗体[EP1809Y] to Nrf2 (phospho S40)

宿主 Rabbit

经测试应用 适用于: Dot blot, ICC/IF, IHC-P, Flow Cyt (Intra), WB

不适用干: IP

种属反应性 与反应: Human

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

(Peptide available as ab133404)

阳性对照 WB: HepG2 whole cell lysate (ab7900). IHC-P: Human tonsil, breast carcinoma, ovarian

carcinoma and cervical carcinoma tissue. ICC/IF: HepG2 cells. Flow Cyt (intra): Jurkat cells.

常规说明 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**.

Rat: We have preliminary internal testing data to indicate this antibody may not react with this

species. Please contact us for more information.

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

克隆 单克隆

克隆编号 EP1809Y

同种型 IgG

应用

The Abpromise guarantee Abpromise™承诺保证使用ab76026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
Dot blot		Use at an assay dependent concentration.
ICC/IF	*** <u>*</u>	1/100 - 1/250.
IHC-P	★★★★☆ (3)	1/100 - 1/500. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. See IHC antigen retrieval protocols.
Flow Cyt (Intra)		1/80 - 1/100. ab172730 - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
WB	★★★★ <u>(13)</u>	1/5000 - 1/50000. Predicted molecular weight: 68 kDa.

应用说明 Is unsuitable for IP.

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.

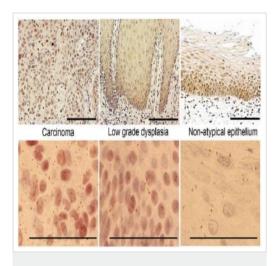
翻译后修饰 Phosphorylation of Ser-40 by PKC in response to oxidative stress dissociates NFE2L2 from its

cytoplasmic inhibitor KEAP1, promoting its translocation into the nucleus.

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

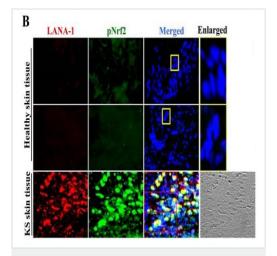
nucleus upon induction by electrophilic agents.

图片



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Nrf2 (phospho S40) antibody [EP1809Y] (ab76026)

Inui et al PLoS One. 2013 Sep 24;8(9):e74398. doi: 10.1371/journal.pone.0074398. eCollection 2013. Fig 2. Reproduced under the Creative Commons license http://creativecommons.org/licenses/by/4.0/



Immunocytochemistry/ Immunofluorescence - Anti-Nrf2 (phospho S40) antibody [EP1809Y] (ab76026)

Giyshi et al PLoS Pathog. 2014 Oct 23;10(10):e1004460. doi: 10.1371/journal.ppat.1004460. eCollection 2014 Oct. Fig 1. Reproduced under the Creative Commons license http://creativecommons.org/licenses/by/4.0/

Nrf2 was abundantly expressed in carcinomas, low grade dysplasias, and non-atypical epithelia of oral tissue.

Representative findings of Nrf2 staining in carcinoma (left), in low grade dysplasia (middle), and in non-atypical epithelium (right).

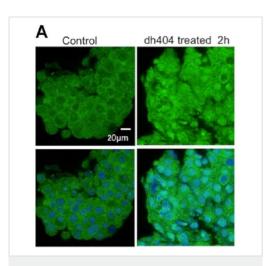
Corresponding PLA signals are displayed in the lower row. Scale bar; 100 µm.

Surgical specimens were transferred to 10% buffered formalin and fixed overnight. The fixed samples were embedded in paraffin, and serially sliced into 5 μ m sections. After dewaxing, sections were autoclaved at 120°C for 1 min in 10 mM sodium citrate buffer (pH 6.0), and immersed in 0.3% H₂O₂. They were then incubated overnight at 4°C with primary antibody to Nrf2 (diluted 1:200). The sections were rinsed with 1×PBS and incubated with the secondary antibody conjugated with horseradish peroxidase at room temperature for 1 hour. The sections were then stained with 3.3′-diaminobenzidinetetrahydrochloride (DAB) and counterstained with hematoxylin.

Immunofluorescence analysis of Nrf2 levels in Kaposi's sarcoma skin lesions.

B) Healthy skin (top two rows) and KS skin tissue (bottom row) were double-stained for LANA-1 (Alexa-Fluor 594- red) and host phosphorylated pNrf2 (ab76026) (Alexa-Fluor[®]488 – green). DAPI was used to visualize the nuclei, and the triple merge of LANA-1, pNrf2 and DAPI is shown in the third column.

Yellow square=enlarged area.

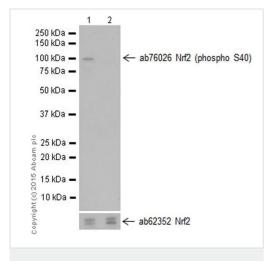


Immunocytochemistry/ Immunofluorescence - Anti-Nrf2 (phospho S40) antibody [EP1809Y] (ab76026)

Masuda et al PLoS One. 2015 Jun 25;10(6):e0131012. doi: 10.1371/journal.pone.0131012. eCollection 2015. Fig 1. Reproduced under the Creative Commons license http://creativecommons.org/licenses/by/4.0/

Nrf2 Translocation from cytoplasm to nucleus.

(A) Human islets were treated with dh404 for 0.5, 1 or 2 hours. The treated and untreated samples were stained with Nrf2 antibody ab76026 (Green) and DAPI (Blue). The con-focal microscope clearly showed that the Nrf2 translocation from cytoplasm to nucleus in the dh404 treated human islet cells



Western blot - Anti-Nrf2 (phospho S40) antibody [EP1809Y] (ab76026) All lanes: Anti-Nrf2 (phospho S40) antibody [EP1809Y] (ab76026)

at 1/50000 dilution (purified)

Lane 1: untreated HepG2 cell lysate

Lane 2: HepG2 treated with phosphatase lysate

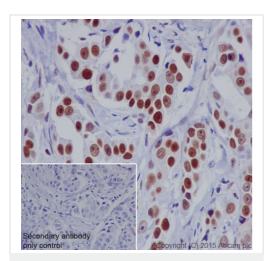
Lysates/proteins at 10 µg per lane.

Secondary

All lanes : HRP goat anti-rabbit lgG (H+L) at 1/20000 dilution

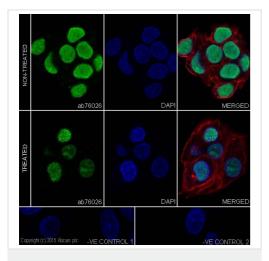
Predicted band size: 68 kDa **Observed band size:** 90 kDa

Blocking buffer: 5% NFDM/TBST Dilution buffer: 5% NFDM/TBST



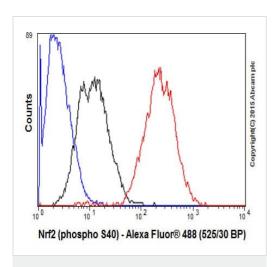
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Nrf2 (phospho S40) antibody [EP1809Y] (ab76026)

Immunohistochemical staining of paraffin embedded human breast carcinoma with purified ab76026 at a working dilution of 1/500. The secondary antibody used is HRP goat anti-rabbit lgG H&L (ab97051) at 1/500. The sample is counter-stained with hematoxylin. Antigen retrieval was performed using Tris-EDTA buffer, pH 9.0. PBS was used instead of the primary antibody as the negative control, and is shown in the inset.

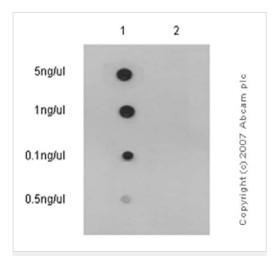


Immunocytochemistry/ Immunofluorescence - Anti-Nrf2 (phospho S40) antibody [EP1809Y] (ab76026)

Immunofluorescence staining of HepG2 cells with purified ab76026 at a working dilution of 1/100, counter-stained with DAPI. The treated cells were treated with alkaline phosphatase for 1 h at 37°C. The secondary antibody was Alexa Fluor[®] 488 goat anti-rabbit (ab150077), used at a dilution of 1/1000. ab7291, a mouse antitubulin antibody (1/1000), was used to stain tubulin along with ab150120 (Alexa Fluor[®] 594 goat anti-mouse, 1/1000), shown in the top right hand panel. The cells were fixed in 4% PFA and permeabilized using 0.1% Triton X 100. The negative controls are shown in bottom middle and right hand panels - for negative control 1, purified ab76026 was used at a dilution of 1/500 followed by an Alexa Fluor[®] 594 goat anti-mouse antibody (ab150120) at a dilution of 1/500. For negative control 2, ab7291 (mouse antitubulin) was used at a dilution of 1/500 followed by an Alexa Fluor[®] 488 goat anti-rabbit antibody (ab150077) at a dilution of 1/400.



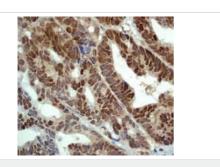
Flow Cytometry (Intracellular) - Anti-Nrf2 (phospho S40) antibody [EP1809Y] (ab76026) Overlay histogram showing Jurkat cells fixed in 4% PFA and stained with purified ab76026 at a dilution of 1 in 80 (red line). The secondary antibody used was FITC goat anti-rabbit at a dilution of 1 in 500. Rabbit monoclonal lgG was used as an isotype control (black line) and cells incubated in the absence of both primary and secondary antibody were used as a negative control (blue line).



Dot Blot - Anti-Nrf2 (phospho S40) antibody [EP1809Y] (ab76026) Dot blot analysis of Nrf2 peptides using unpurified ab76026 at 1/1000 dilution followed by Goat Anti-Rabbit IgG, (H+L),Peroxidase conjugated secondary antibody at 1/1000 dilution. Blocking and diluting buffer was 5% NFDM/TBST.

Lane 1: Nrf2 (pS40) phospho peptide

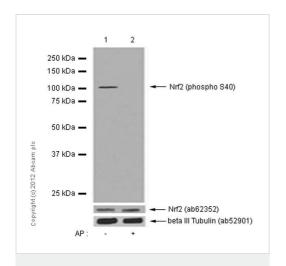
Lane 2: Nrf2 non-phospho peptide



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Nrf2 (phospho S40) antibody [EP1809Y] (ab76026)

Immunohistochemical analysis of paraffin-embedded human breast carcinoma using unpurified ab76026 at 1/100 dilution.

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Western blot - Anti-Nrf2 (phospho S40) antibody [EP1809Y] (ab76026)

All lanes : Anti-Nrf2 (phospho S40) antibody [EP1809Y] (ab76026) at 1/10000 dilution (unpurified)

Lane 1 : Untreated HepG2 (human hepatocellular carcinoma) whole cell lysates 20µg

Lane 2: HepG2 (human hepatocellular carcinoma) treated with Alkaline Phosphatase (AP) whole cell lysates 20µg.

Secondary

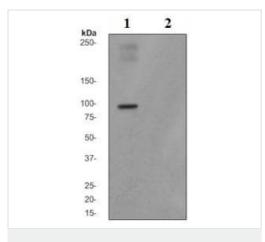
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/1000 dilution

Predicted band size: 68 kDa

Observed band size: 100 kDa

Blocking buffer: 5% NFDM/TBST, dilution buffer: 5% NFDM /TBST,

exposure time: 15 seconds



Western blot - Anti-Nrf2 (phospho S40) antibody [EP1809Y] (ab76026)

All lanes : Anti-Nrf2 (phospho S40) antibody [EP1809Y] (ab76026) at 1/20000 dilution (unpurified)

Lane 1: HepG2 cell lysate

Lane 2: HepG2 cell lysate treated with AP

Lysates/proteins at 10 µg per lane.

Secondary

All lanes: HRP labelled goat anti-rabbit at 1/1000 dilution

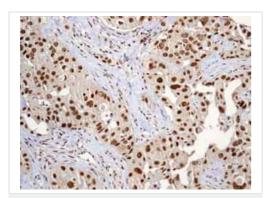
Predicted band size: 68 kDa **Observed band size:** 90 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Nrf2 (phospho S40) antibody [EP1809Y] (ab76026)

This image is courtesy of an anonymous Abreview

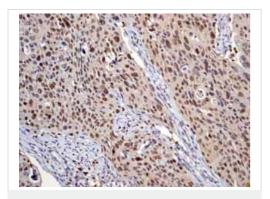
Unpurified ab76026 staining Nrf2 (phospho S40) in Human normal lung tissue sections by IHC-P (Formaldehyde-fixed paraffinembedded sections). Tissue was fixed with formaldehyde and blocked with 1% casein for 30 minutes at 4°C. Antigen retrieval was by heat mediation. Samples were incubated with primary antibody (1/50) in 1% casein for 24 hours at 4°C. An undiluted HRP-conjugated Goat polyclonal to rabbit IgG was used as the secondary antibody.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Nrf2 (phospho S40) antibody [EP1809Y] (ab76026)

Unpurified ab76026 showing positive staining in Breast carcinoma tissue.

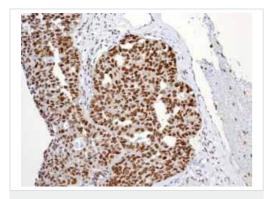
Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Nrf2 (phospho S40) antibody [EP1809Y] (ab76026)

Unpurified ab76026 showing positive staining in Cervical carcinoma tissue.

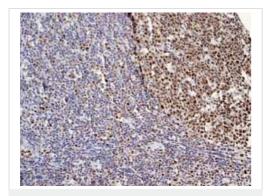
Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Nrf2 (phospho S40) antibody [EP1809Y] (ab76026)

Unpurified ab76026 showing positive staining in Ovarian carcinoma tissue.

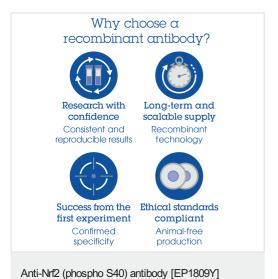
Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Nrf2 (phospho S40) antibody [EP1809Y] (ab76026)

Unpurified ab76026 showing positive staining in Normal tonsil tissue.

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



(ab76026)

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