

Anti-Phospho-Nrf2 (S40) NFE2L2 Rabbit Monoclonal Antibody

Catalog # ABO13121

Specification

Anti-Phospho-Nrf2 (S40) NFE2L2 Rabbit Monoclonal Antibody - Product Information

Application WB, IHC, IF, ICC, IP, FC

Primary Accession
Host
Rabbit
Isotype
Rabbit IgG

Reactivity Rat, Human, Mouse

Clonality Monoclonal Format Liquid

Description

Anti-Phospho-Nrf2 (S40) NFE2L2 Rabbit Monoclonal Antibody . Tested in WB, IHC, ICC/IF, IP, Flow Cytometry applications. This antibody reacts with Human, Mouse, Rat.

Anti-Phospho-Nrf2 (S40) NFE2L2 Rabbit Monoclonal Antibody - Additional Information

Gene ID 4780

Other Names

Nuclear factor erythroid 2-related factor 2, NF-E2-related factor 2, NFE2-related factor 2, Nrf-2, HEBP1, Nuclear factor, erythroid derived 2, like 2, NFE2L2 {ECO:0000303|PubMed:29018201, ECO:0000312|HGNC:HGNC:7782}

Calculated MW 67827 MW KDa

Application Details

WB 1:1000-1:2000
IHC 1:50-1:200
ICC/IF 1:50-1:200
IP 1:50
FC 1:50</br>

Subcellular Localization

Cytoplasm, cytosol. Nucleus. Cytosolic under unstressed conditions, translocates into the nucleus upon induction by electrophilic agents.

Tissue Specificity

Widely expressed. Highest expression in adult muscle, kidney, lung, liver and in fetal muscle.

Contents

Rabbit IgG in phosphate buffered saline, pH 7.4, 150mM NaCl, 0.02% sodium azide and 50% glycerol, 0.4-0.5mg/ml BSA.

Immunogen

A synthesized peptide derived from human Phospho-Nrf2 (S40)

Purification

Affinity-chromatography

Storage Store at -20°C for one year. For short term



storage and frequent use, store at 4°C for up to one month. Avoid repeated freeze-thaw cycles.

Anti-Phospho-Nrf2 (S40) NFE2L2 Rabbit Monoclonal Antibody - Protein Information

Name NFE2L2 {ECO:0000303|PubMed:29018201, ECO:0000312|HGNC:HGNC:7782}

Function

Transcription factor that plays a key role in the response to oxidative stress: binds to antioxidant response (ARE) elements present in the promoter region of many cytoprotective genes, such as phase 2 detoxifying enzymes, and promotes their expression, thereby neutralizing reactive electrophiles (PubMed:11035812, PubMed:19489739, PubMed:29018201, PubMed:31398338). In normal conditions, ubiquitinated and degraded in the cytoplasm by the BCR(KEAP1) complex (PubMed:11035812, PubMed:15601839, PubMed:29018201). In response to oxidative stress, electrophile metabolites inhibit activity of the BCR(KEAP1) complex, promoting nuclear accumulation of NFE2L2/NRF2, heterodimerization with one of the small Maf proteins and binding to ARE elements of cytoprotective target genes (PubMed: 19489739, PubMed:29590092). The NFE2L2/NRF2 pathway is also activated in response to selective autophagy: autophagy promotes interaction between KEAP1 and SQSTM1/p62 and subsequent inactivation of the BCR(KEAP1) complex, leading to NFE2L2/NRF2 nuclear accumulation and expression of cytoprotective genes (PubMed:20452972). The NFE2L2/NRF2 pathway is also activated during the unfolded protein response (UPR), contributing to redox homeostasis and cell survival following endoplasmic reticulum stress (By similarity). May also 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 (PubMed:7937919). Also plays an important role in the regulation of the innate immune response and antiviral cytosolic DNA sensing. It is a critical regulator of the innate immune response and survival during sepsis by maintaining redox homeostasis and restraint of the dysregulation of pro-inflammatory signaling pathways like MyD88- dependent and -independent and TNF-alpha signaling (By similarity). Suppresses macrophage inflammatory response by blocking pro- inflammatory cytokine transcription and the induction of IL6 (By similarity). Binds to the proximity of pro-inflammatory genes in macrophages and inhibits RNA Pol II recruitment. The inhibition is independent of the NRF2-binding motif and reactive oxygen species level (By similarity). Represses antiviral cytosolic DNA sensing by suppressing the expression of the adapter protein STING1 and decreasing responsiveness to STING1 agonists while increasing susceptibility to infection with DNA viruses (PubMed: 30158636). Once activated, limits the release of pro-inflammatory cytokines in response to human coronavirus SARS-CoV-2 infection and to virus-derived ligands through a mechanism that involves inhibition of IRF3 dimerization. Also inhibits both SARS-CoV-2 replication, as well as the replication of several other pathogenic viruses including Herpes Simplex Virus-1 and-2, Vaccinia virus, and Zika virus through a type I interferon (IFN)- independent mechanism (PubMed:33009401).

Cellular Location

Cytoplasm, cytosol. Nucleus {ECO:0000255|PROSITE-ProRule:PRU00978,



ECO:0000269|PubMed:11035812, ECO:0000269|PubMed:15601839, ECO:0000269|PubMed:21196497, ECO:0000269|PubMed:29983246}. Note=Cytosolic under unstressed conditions: ubiquitinated and degraded by the BCR(KEAP1) E3 ubiquitin ligase complex (PubMed:15601839, PubMed:21196497). Translocates into the nucleus upon induction by electrophilic agents that inactivate the BCR(KEAP1) E3 ubiquitin ligase complex (PubMed:21196497)

Tissue Location

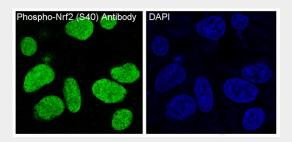
Widely expressed. Highest expression in adult muscle, kidney, lung, liver and in fetal muscle

Anti-Phospho-Nrf2 (S40) NFE2L2 Rabbit Monoclonal Antibody - Protocols

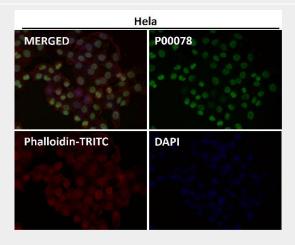
Provided below are standard protocols that you may find useful for product applications.

- Western Blot
- Blocking Peptides
- Dot Blot
- <u>Immunohistochemistry</u>
- Immunofluorescence
- Immunoprecipitation
- Flow Cytomety
- Cell Culture

Anti-Phospho-Nrf2 (S40) NFE2L2 Rabbit Monoclonal Antibody - Images

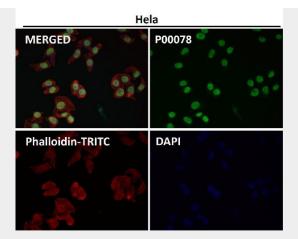


Immunofluorescent analysis of HepG2 cells, using Phospho-Nrf2 (S40) Antibody.

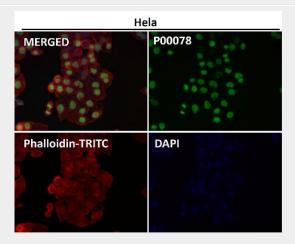


Immunofluorescent analysis using the Antibody at 1:50 dilution.





Immunofluorescent analysis using the Antibody at 1:150 dilution.



Immunofluorescent analysis using the Antibody at 1:500 dilution.

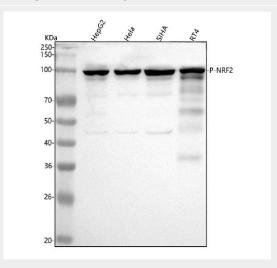


Figure 1. Western blot analysis of Nrf2 using anti-Nrf2 antibody (P00078).

Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions.

- Lane 1: human HepG2 whole cell lysates,
- Lane 2: human Hela whole cell lysates,
- Lane 3: human SiHa whole cell lysates,
- Lane 4: human RT4 whole cell lysates.



After electrophoresis, proteins were transferred to a nitrocellulose membrane at 150 mA for 50-90 minutes. Blocked the membrane with 5% non-fat milk/TBS for 1.5 hour at RT. The membrane was incubated with rabbit anti-Nrf2 antigen affinity purified monoclonal antibody (Catalog # P00078) at 1:1000 overnight at 4°C , then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-rabbit IgG-HRP secondary antibody at a dilution of 1:500 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1002) with Tanon 5200 system. A specific band was detected for Nrf2 at approximately 100 kDa. The expected band size for Nrf2 is at 68 kDa.

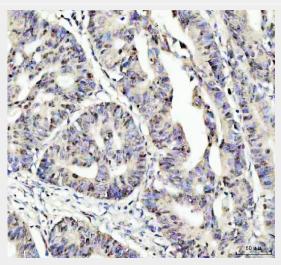


Figure 2. IHC analysis of Nrf2 using anti-Nrf2 antibody (P00078).

Nrf2 was detected in a paraffin-embedded section of human colorectal adenocarcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1:50 rabbit anti-Nrf2 Antibody (P00078) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.

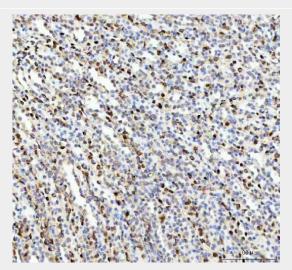


Figure 3. IHC analysis of Nrf2 using anti-Nrf2 antibody (P00078).

Nrf2 was detected in a paraffin-embedded section of mouse kidney tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1:50 rabbit anti-Nrf2 Antibody (P00078) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed



using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.

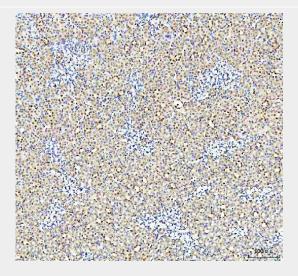


Figure 4. IHC analysis of Nrf2 using anti-Nrf2 antibody (P00078).

Nrf2 was detected in a paraffin-embedded section of rat kidney tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1:50 rabbit anti-Nrf2 Antibody (P00078) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.