Anti-Nrf2 Antibody [PSH11-20]

HA723302

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, ChIP
Molecular Wt:	Predicted band size: 68 kDa
Clone number:	PSH11-20

Description: Nuclear factor erythroid 2-related factor 2 (NRF2), also known as nuclear factor erythroid-derived 2-like 2, is a transcription factor that in humans is encoded by the NFE2L2 gene. NRF2 is a basic leucine zipper (bZIP) protein that may regulate the expression of antioxidant proteins that protect against oxidative damage triggered by injury and inflammation, according to preliminary research. In vitro, NRF2 binds to antioxidant response elements (AREs) in the promoter regions of genes encoding cytoprotective proteins. NRF2 induces the expression of heme oxygenase 1 in vitro leading to an increase in phase II enzymes. NRF2 also inhibits the NLRP3 inflammasome. NRF2 appears to participate in a complex regulatory network and performs a pleiotropic role in the regulation of metabolism, inflammation, autophagy, proteostasis, mitochondrial physiology, and immune responses. Several drugs that stimulate the NFE2L2 pathway are being studied for treatment of diseases that are caused by oxidative stress.

Immunogen: Recombinant protein within human Nrf2 aa 51-350.

Positive control: MEF treated with 10μM MG-132 for 8 hours cell lysate, HeLa treated with 2μM MG-132 for 18 hours cell lysate, C6 treated with 25μM MG-132 for 4 hours cell lysate, MEF cells treated with 2μM MG-132 for 18 hours, HCT 116 cells treated with 25μM MG-132 for 4 hours.

Subcellular location: Cytoplasm, cytosol, Nucleus.

Database links: SwissProt: Q16236 Human | Q60795 Mouse | O54968 Rat

Recommended Dilutions:

WB	1:500-1:2,000
IF-Cell	1:100
ChIP	Use 0.5~2 μg for 25 μg of chromatin.

Storage Buffer: PBS (pH7.4), 0.1% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

Storage Instruction: Store at +4℃ after thawing. Aliquot store at -20℃. Avoid repeated freeze / thaw cycles.

Purity: Protein A affinity purified.

Images

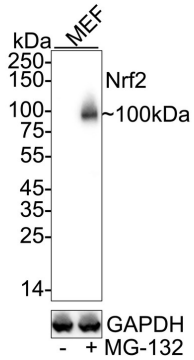

Fig1: Western blot analysis of Nrf2 on different lysates with Rabbit anti-Nrf2 antibody (HA723302) at 1/2,000 dilution.

Lane 1: MEF cell lysate
Lane 2: MEF treated with 10μM MG-132 for 8 hours cell lysate

Lysates/proteins at 20 μg/Lane.

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

Exposure time: 14 seconds; ECL: K1801;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (HA723302) at 1/2,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

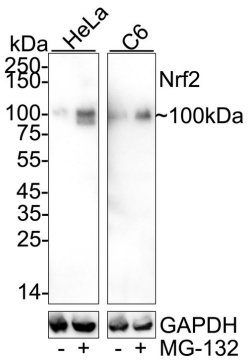

Fig2: Western blot analysis of Nrf2 on different lysates with Rabbit anti-Nrf2 antibody (HA723302) at 1/2,000 dilution.

Lane 1: HeLa cell lysate
Lane 2: HeLa treated with 2μM MG-132 for 18 hours cell lysate
Lane 3: C6 cell lysate
Lane 4: C6 treated with 25μM MG-132 for 4 hours cell lysate

Lysates/proteins at 20 μg/Lane.

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

Exposure time: 2 minutes; ECL: K1801;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (HA723302) at 1/2,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

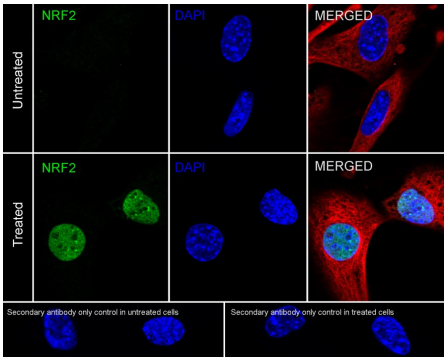

Fig3: Immunocytochemistry analysis of MEF cells untreated / treated with 2μM MG-132 for 18 hours labeling Nrf2 with Rabbit anti-Nrf2 antibody (HA723302) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-Nrf2 antibody (HA723302) at 1/100 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (HA601187, red) was stained at 1/100 dilution overnight at +4℃. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

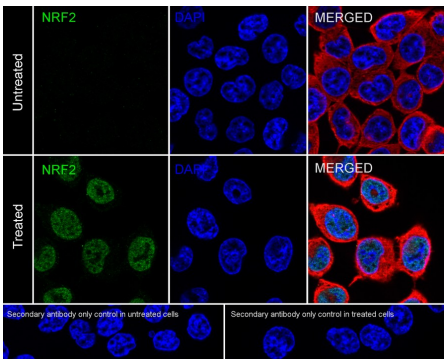

Fig4: Immunocytochemistry analysis of HCT 116 cells untreated / treated with 25μM MG-132 for 4 hours labeling Nrf2 with Rabbit anti-Nrf2 antibody (HA723302) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-Nrf2 antibody (HA723302) at 1/100 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (HA601187, red) was stained at 1/100 dilution overnight at +4℃. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

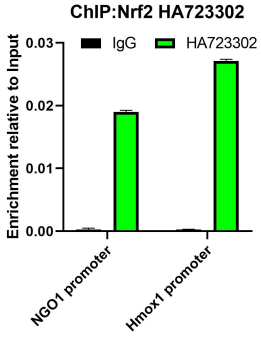

Fig5: Chromatin immunoprecipitations were performed with cross-linked chromatin from MEF cells treated with 10μM MG-132 for 8 hours with Nrf2 (HA723302) or Normal Rabbit IgG according to the ChIP protocol. The enriched DNA was quantified by real-time PCR using indicated primers. The amount of immunoprecipitated DNA in each sample is represented as signal relative to the total amount of input chromatin, which is equivalent to one.

Background References

1. He F et al. NRF2, a Transcription Factor for Stress Response and Beyond. Int J Mol Sci. 2020 Jul
2. Ulasov AV et al. Nrf2/Keap1/ARE signaling: Towards specific regulation. Life Sci. 2022 Feb