#12721 Store at -20°C

NRF2 (D1Z9C) XP® Rabbit mAb

Cell Signaling TECHNOLOGY®

☐ Small 100 µl (10 western blots)
☐ Petite 40 µl (4 western blots)

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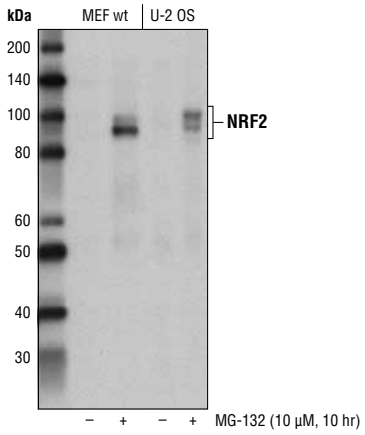
Entrez-Gene ID # 4780
UniProt ID #Q16236

Applications	Species Cross-Reactivity*	Molecular Wt.	Isotype
W, IP, ChIP, F Endogenous	H, M, Mk	97–100 kDa	Rabbit IgG**

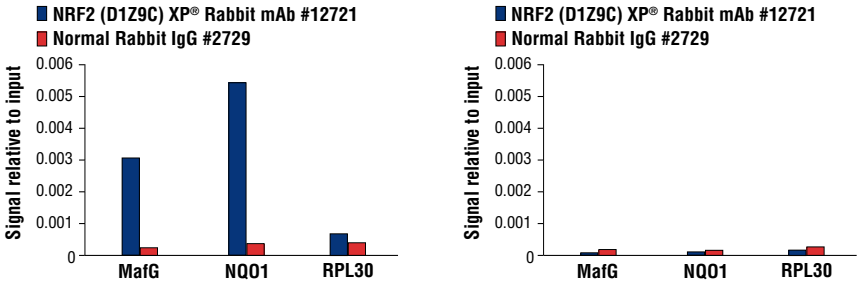
Background: The nuclear factor-like 2 (NRF2) transcriptional activator binds antioxidant response elements (ARE) of target gene promoter regions to regulate expression of oxidative stress response genes. Under basal conditions, the NRF2 inhibitor INrf2 (also called KEAP1) binds and retains NRF2 in the cytoplasm where it can be targeted for ubiquitin-mediated degradation (1). Small amounts of constitutive nuclear NRF2 maintain cellular homeostasis through regulation of basal expression of antioxidant response genes. Following oxidative or electrophilic stress, KEAP1 releases NRF2, thereby allowing the activator to translocate to the nucleus and bind to ARE-containing genes (2). The coordinated action of NRF2 and other transcription factors mediates the response to oxidative stress (3). Altered expression of NRF2 is associated with chronic obstructive pulmonary disease (COPD) (4). NRF2 activity in lung cancer cell lines directly correlates with cell proliferation rates, and inhibition of NRF2 expression by siRNA enhances anti-cancer drug-induced apoptosis (5).

Specificity/Sensitivity: NRF2 (D1Z9C) XP® Rabbit mAb recognizes endogenous levels of total NRF2 protein.

Source/Purification: Monoclonal antibody is produced by immunizing animals with a synthetic peptide corresponding to residues surrounding Ala275 of human NRF2 protein.



Western blot analysis of extracts from MEF wt and U-2 OS cells, untreated (-) or treated with MG-132 #2194 (10 µM, 10 hr; +), using NRF2 (D1Z9C) XP® Rabbit mAb.



Chromatin immunoprecipitations were performed with cross-linked chromatin from 4 x 10^6 MEF NRF2 wild-type (left) and NRF2 knock-out (right) cells, both treated with DEM (50 µM, 3 hr), and 5 µl of NRF2 (D1Z9C) XP® Rabbit mAb or 2 µl of Normal Rabbit IgG #2729 using SimpleChIP® Enzymatic Chromatin IP Kit (Magnetic Beads) #9003. The enriched DNA was quantified by real-time PCR using mouse MafG intron 1 primers, SimpleChIP® Mouse NQO1 Promoter Primers #12635, and SimpleChIP® Mouse RPL30 Intron 2 Primers #7015. 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.

IMPORTANT: For western blots, incubate membrane with diluted antibody in 5% w/v nonfat dry milk, 1X TBS, 0.1% Tween® 20 at 4°C with gentle shaking, overnight.

Storage: Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA, 50% glycerol and less than 0.02% sodium azide. Store at –20°C. *Do not aliquot the antibody.*

***Species cross-reactivity is determined by western blot.**

****Anti-rabbit secondary antibodies must be used to detect this antibody.**

Recommended Antibody Dilutions:

Western blotting	1:1000
Immunoprecipitation	1:50
Chromatin IP	1:100
Flow Cytometry	1:6400

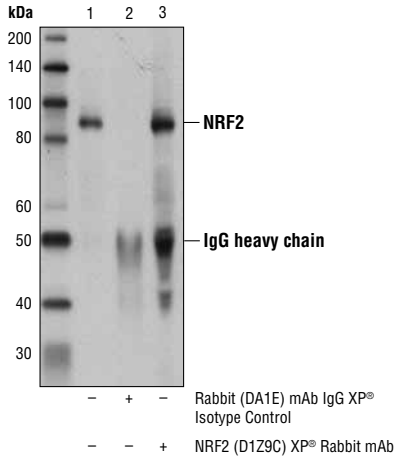
For product specific protocols please see the web page for this product at www.cellsignal.com.

Please visit www.cellsignal.com for a complete listing of recommended companion products.

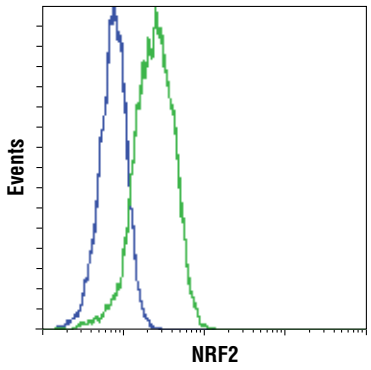
Background References:

(1) Cullinan, S.B. et al. (2004) *Mol Cell Biol* 24, 8477-86.
(2) Nguyen, T. et al. (2005) *J Biol Chem* 280, 32485-92.
(3) Jaiswal, A.K. (2004) *Free Radic Biol Med* 36, 1199-207.
(4) Suzuki, M. et al. (2008) *Am J Respir Cell Mol Biol* 39, 673-82.
(5) Homma, S. et al. (2009) *Clin Cancer Res* 15, 3423-32.

Applications Key: **W**—Western **IP**—Immunoprecipitation **IHC**—Immunohistochemistry **ChIP**—Chromatin Immunoprecipitation **IF**—Immunofluorescence **F**—Flow cytometry **E-P**—ELISA-Peptide
Species Cross-Reactivity Key: **H**—human **M**—mouse **R**—rat **Hm**—hamster **Mk**—monkey **Mi**—mink **C**—chicken **Dm**—D. melanogaster **X**—Xenopus **Z**—zebrafish **B**—bovine **Dg**—dog **Pg**—pig **Sc**—S. cerevisiae **Ce**—C. elegans **Hr**—horse **All**—all species expected Species enclosed in parentheses are predicted to react based on 100% homology.



Immunoprecipitation of NRF2 from MEF wt cell extracts treated with MG-132 #2194 (10 µM, 10 hr) using Rabbit (DA1E) mAb IgG XP® Isotype Control #3900 (lane 2) or NRF2 (D1Z9C) XP® Rabbit mAb (lane 3). Lane 1 is 10% input. Western blot analysis was performed using NRF2 (D1Z9C) XP® Rabbit mAb (lane 3).



Flow cytometric analysis of MEF wt cells, untreated (blue) and treated with MG-132 #2194 (green), using NRF2 (D1Z9C) XP® Rabbit mAb. Anti-rabbit IgG (H+L), F(ab')$_2$ Fragment (Alexa Fluor® 647 Conjugate) #4414 was used as a secondary antibody.