abcam

Product datasheet

Anti-NRF1 antibody [EPR5554(N)] - ChIP Grade - BSA and Azide free ab221792

Recombinant RabMAb

2 References 14 Images

Overview

Product name	Anti-NRF1 antibody [EPR5554(N)] - ChIP Grade - BSA and Azide free
Description	Rabbit monoclonal [EPR5554(N)] to NRF1 - ChIP Grade – BSA and Azide free
Host species	Rabbit
Tested applications	**Suitable for:** WB, IHC-P, ICC/IF, IP, Flow Cyt (Intra), ChIP, ChIC/CUT&RUN-seq, ChIP-sequencing
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: MCF-7, HeLa and 293T cell lysates and human fetal heart, mouse heart, mouse brain, rat heart and rat brain tissue lysates. IHC-P: Human gastric adenocarcinoma, human cervical carcinoma and human skeletal muscle tissues. ICC/IF: HeLa and MCF-7 cells. Flow Cyt (intra): 293T cells. IP: 293T cell lysate. ChIP-Seq: HeLA cells.
General notes	ab221792 is the carrier-free version of **ab175932**. Our **carrier-free** antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency. This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications. Use our **conjugation kits** for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold. This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc. 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**.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C. Do Not Freeze.
Storage buffer	pH: 7.20 Constituent: PBS
Carrier free	Yes
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR5554(N)
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab221792 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

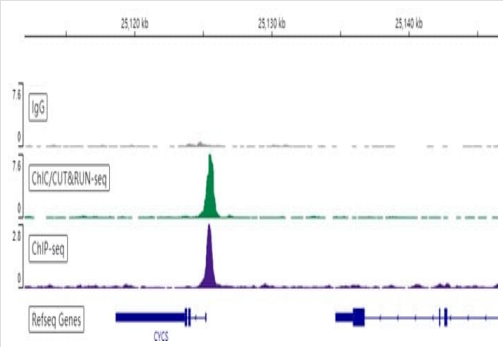
Application	Abreviews	Notes
WB		Use at an assay dependent concentration. Predicted molecular weight: 54 kDa.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. See **IHC antigen retrieval protocols**.
ICC/IF		Use at an assay dependent concentration.
IP		Use at an assay dependent concentration.
Flow Cyt (Intra)		Use at an assay dependent concentration. **ab199376**- Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
ChIP		Use at an assay dependent concentration.
ChIC/CUT&RUN-seq		Use at an assay dependent concentration.
ChIP-sequencing		Use 8µg for 10^7 cells.

Target

Function Transcription factor that activates the expression of the EIF2S1 (EIF2-alpha) gene. Links the

	transcriptional modulation of key metabolic genes to cellular growth and development. Implicated in the control of nuclear genes required for respiration, heme biosynthesis, and mitochondrial DNA transcription and replication.
Tissue specificity	Ubiquitously expressed with strongest expression in skeletal muscle.
Sequence similarities	Belongs to the NRF1/Ewg family.
Post-translational modifications	Phosphorylation enhances DNA binding.
Cellular localization	Nucleus.

Images



ChIC/CUT&RUN sequencing - Anti-NRF1 antibody [EPR5554(N)] - ChIP Grade - BSA and Azide free (ab221792)

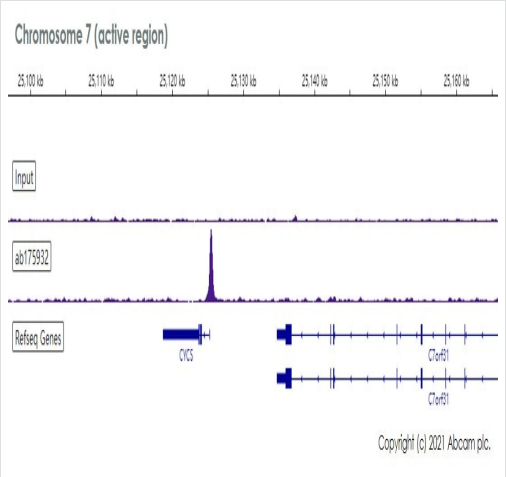
ChIC/CUT&RUN was performed using a pAG-MNAse at a final concentration of 700 ng/µL, 2 x 10^5 HeLa (Human cervix adenocarcinoma epithelial cell line) cells and 5 µg of **ab175932** [EPR5554(N)]. The resulting DNA was sequenced on the Illumina NovaSeq 6000 to a depth of 10 million reads. The negative IgG control **ab172730** is also shown.

The ChIP data was conducted on chromatin prepared from HeLa cells. Cells were fixed with 1% formaldehyde for 10 minutes. ChIP was performed with 10^7 HeLa cells and 8 µg of **ab175932**. ChIP DNA was sequenced on the Illumina NovaSeq 6000 to a depth of 30 million reads.

Additional screenshots of mapped reads can be downloaded **here**.

The University of Geneva owns patents relevant to ChIC (Chromatin Immuno-Cleavage) methods.

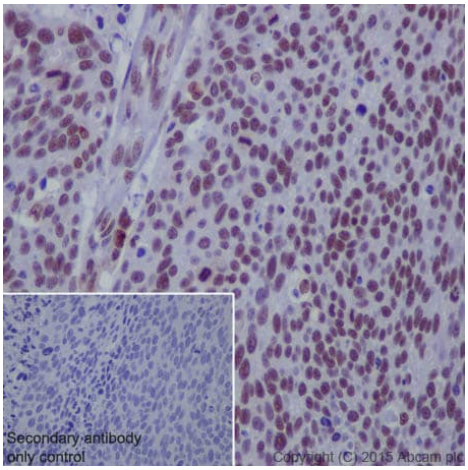
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab175932**).



ChIP-sequencing - Anti-NRF1 antibody [EPR5554(N)] - ChIP Grade - BSA and Azide free (ab221792)

Chromatin was prepared from HeLa cells. Cells were fixed with 1% formaldehyde for 10 minutes. ChIP was performed with 10^7 HeLa cells and 8 µg of **ab175932** [EPR5554(N)]. ChIP DNA was sequenced on the Illumina NovaSeq 6000 to a depth of 30 million reads.

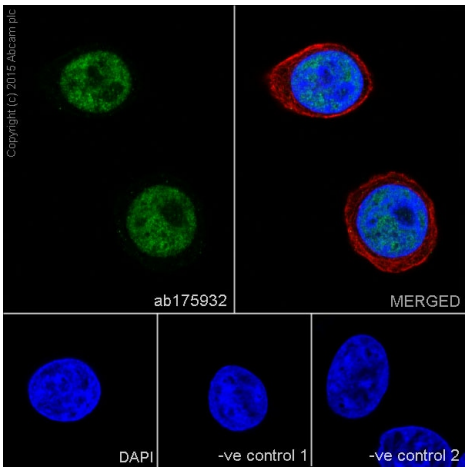
Additional screenshots of mapped reads can be downloaded **here**.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NRF1 antibody [EPR5554(N)] - ChIP Grade - BSA and Azide free (ab221792)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human cevical carcinoma tissue labelling NRF1 with purified **ab175932** at a dilution of 1/100. Heat mediated antigen retrieval was performed using EDTA buffer pH 9. **ab97051**, a HRP-conjugated goat anti-rabbit IgG (H+L) was used as the secondary antibody (1/500). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab175932**).



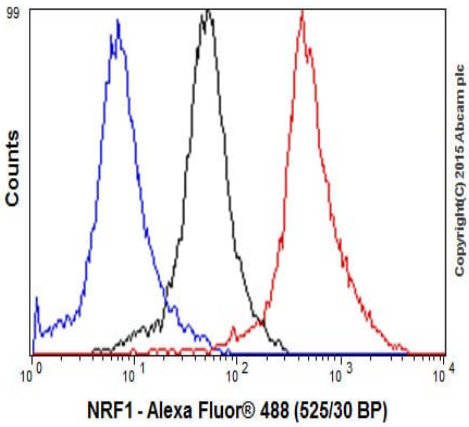
Immunocytochemistry/ Immunofluorescence - Anti-NRF1 antibody [EPR5554(N)] - ChIP Grade - BSA and Azide free (ab221792)

Immunocytochemistry/Immunofluorescence analysis of MCF-7 cells labelling NRF1 with purified **ab175932** at a dilution of 1/100. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. **ab150077**, an Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/1000) was used as the secondary antibody. DAPI (blue) was used as the nuclear counterstain. **ab7291**, a mouse anti-tubulin (1/1000) and **ab150120**, an Alexa Fluor® 594-conjugated goat anti-mouse IgG (1/1000) were also used.

Control 1: primary antibody (1/100) and secondary antibody, **ab150120**, an Alexa Fluor® 594-conjugated goat anti-mouse IgG (1/1000).

Control 2: **ab7291** (1/1000) and secondary antibody, **ab150077**, an Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/1000).

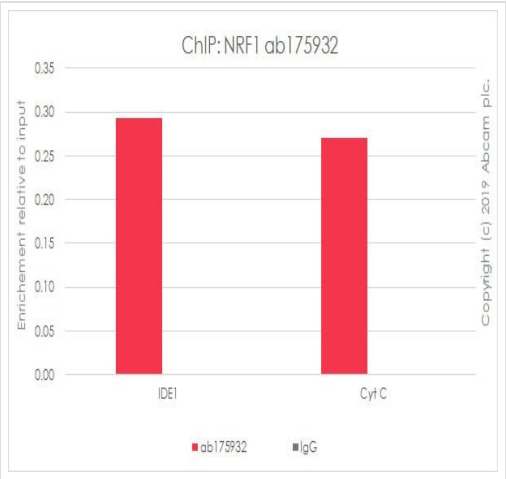
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab175932**).



Flow Cytometry (Intracellular) - Anti-NRF1 antibody [EPR5554(N)] - ChIP Grade - BSA and Azide free (ab221792)

Intracellular Flow Cytometry analysis of 293T cells labelling NRF1 with purified **ab175932** at a dilution of 1/150 (red). Cells were fixed with 80% methanol. A FITC-conjugated goat anti-rabbit IgG (1/500) was used as the secondary antibody. Black - Isotype control, rabbit monoclonal IgG. Blue - Unlabelled control, cells without incubation with primary and secondary antibodies.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab175932**).



ChIP - Anti-NRF1 antibody [EPR5554(N)] - ChIP Grade - BSA and Azide free (ab221792)

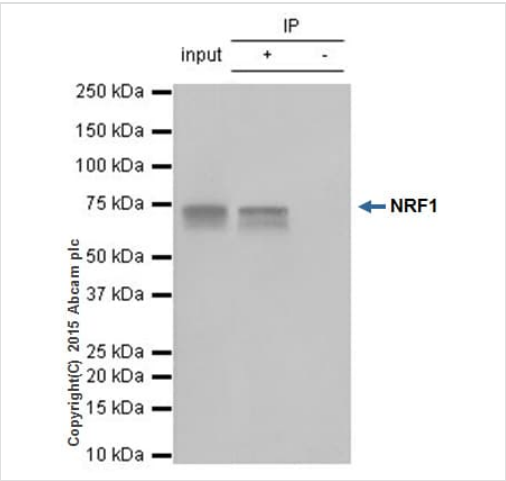
Chromatin was prepared from Hela cells according to the Abcam Dual X-ChIP protocol. Cells were fixed with EGS for 30 minutes, then formaldehyde for 10 minutes.

The ChIP was performed with 25 µg of chromatin, 5 µg of **ab175932** (red), and 20 µl of Protein A/G sepharose beads. 5 µg of rabbit normal IgG was added to the beads control (gray). The immunoprecipitated DNA was quantified by real time PCR (Sybr green approach).

Primers and probes are located in the first kb of the transcribed region.

*http://www.abcam.com/resources?keywords=X%20ChIP%20protocol

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab175932**).



Immunoprecipitation - Anti-NRF1 antibody [EPR5554(N)] - ChIP Grade - BSA and Azide free (ab221792)

ab175932 (purified) at a dilution of 1/50 immunoprecipitating NRF1 in 293T whole cell lysate.

Lane 1 (input): 293T whole cell lysate (10µg)

Lane 2 (+): **ab175932** + 293T whole cell lysate.

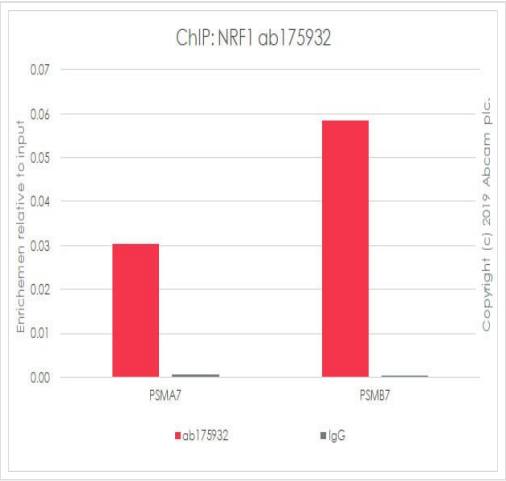
Lane 3 (-): Rabbit monoclonal IgG (**ab172730**) instead of **ab175932** in 293T whole cell lysate.

For western blotting, **ab131366** VeriBlot for IP (HRP) was used for detection at 1/1000 dilution.

Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab175932**).



ChIP - Anti-NRF1 antibody [EPR5554(N)] - ChIP Grade - BSA and Azide free (ab221792)

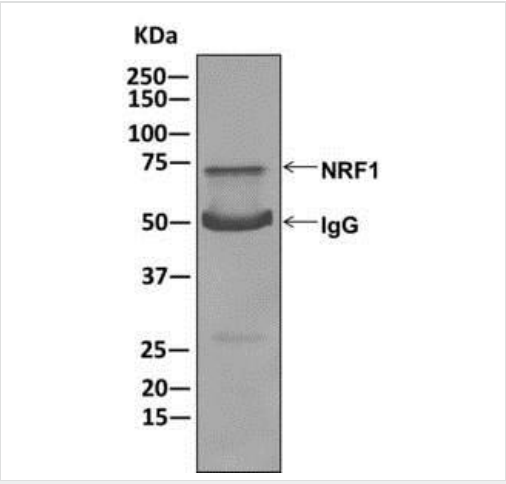
Chromatin was prepared from NIH/3T3 treated with MG-132(2uM 16h) cells according to the Abcam Dual X-ChIP protocol*. Cells were fixed with EGS for 30 minutes, then formaldehyde for 10 minutes.

The ChIP was performed with 25 µg of chromatin, 5 µg of **ab175932** (red), and 20 µl of Protein A/G sepharose beads. 5 µg of rabbit normal IgG was added to the beads control (gray). The immunoprecipitated DNA was quantified by real time PCR (Sybr green approach).

Primers and probes are located in the first kb of the transcribed region.

*http://www.abcam.com/resources?keywords=X%20ChIP%20protocol

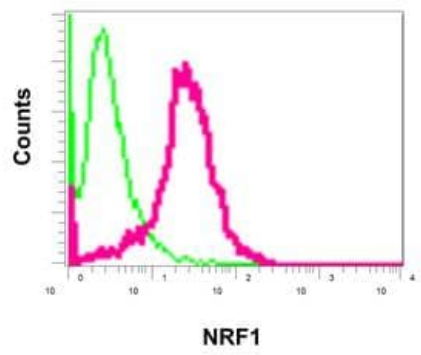
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab175932**).



Immunoprecipitation - Anti-NRF1 antibody [EPR5554(N)] - ChIP Grade - BSA and Azide free (ab221792)

ab175932 (unpurified) at a dilution of 1/10 immunoprecipitating NRF1 in 293T cell lysate.

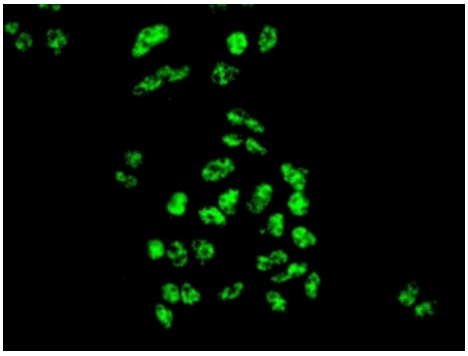
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab175932**).



Flow Cytometry (Intracellular) - Anti-NRF1 antibody [EPR5554(N)] - ChIP Grade - BSA and Azide free (ab221792)

Intracellular flow cytometric analysis of permeabilized 293T cells labeling NRF1 with unpurified **ab175932** at a dilution of 1/10 (red) compared to a negative control (rabbit IgG, green).

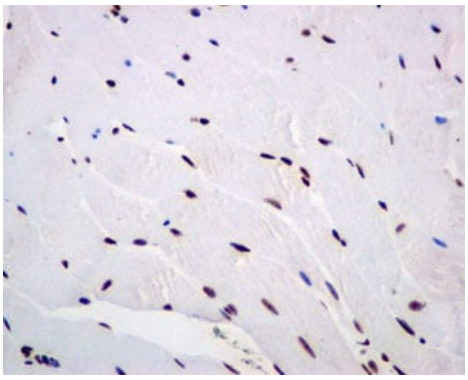
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab175932**).



Immunocytochemistry/ Immunofluorescence - Anti-NRF1 antibody [EPR5554(N)] - ChIP Grade - BSA and Azide free (ab221792)

Immunocytochemistry/Immunofluorescence analysis of HeLa cells labeling NRF1 with unpurified **ab175932** at a dilution of 1/50.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab175932**).

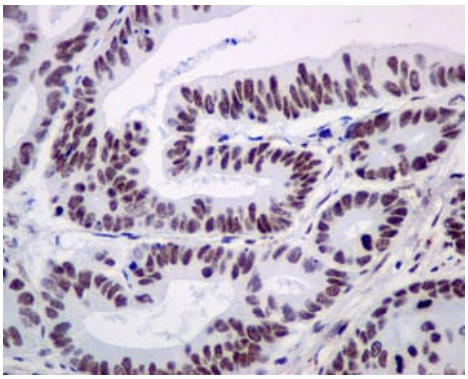


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NRF1 antibody [EPR5554(N)] - ChIP Grade - BSA and Azide free (ab221792)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human skeletal muscle tissue labeling NRF1 with unpurified **ab175932** at a dilution of 1/50.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab175932**).

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

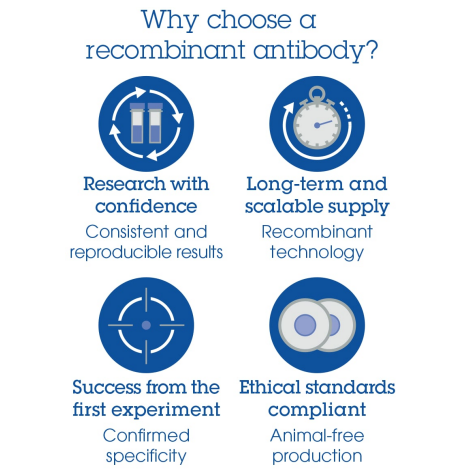


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NRF1 antibody [EPR5554(N)] - ChIP Grade - BSA and Azide free (ab221792)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human gastric adenocarcinoma tissue labeling NRF1 with unpurified **ab175932** at a dilution of 1/50.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab175932**).

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



Anti-NRF1 antibody [EPR5554(N)] - ChIP Grade - BSA and Azide free (ab221792)

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