

A22331

Leader in Biomolecular Solutions for Life Science



[KO Validated] NF-kB p65/RelA Rabbit mAb

Catalog No.: A22331

KO Validated

Recombinant

16 Publications

Basic Information

Observed MW

65kDa

Calculated MW

58kDa/59kDa/60kDa

Category

Monoclonal Antibody

Applications

WB,IHC-P,IF/ICC,IP,ChIP,ELISA

Cross-Reactivity

Human,Mouse,Rat,Monkey

CloneNo number

ARC51088

Background

NF-kappa-B is a ubiquitous transcription factor involved in several biological processes. It is held in the cytoplasm in an inactive state by specific inhibitors. Upon degradation of the inhibitor, NF-kappa-B moves to the nucleus and activates transcription of specific genes. NF-kappa-B is composed of NFKB1 or NFKB2 bound to either REL, RELA, or RELB. The most abundant form of NF-kappa-B is NFKB1 complexed with the product of this gene, RELA. Four transcript variants encoding different isoforms have been found for this gene.

Recommended Dilutions

WB 1:5000 - 1:20000

IHC-P 1:200 - 1:2000

IF/ICC 1:500 - 1:2000

IP 0.5µg-4µg antibody for
200µg-500µg extracts
of whole cells

ChIP 5µg antibody for
10µg-15µg of
Chromatin

ELISA Recommended starting
concentration is 1
µg/mL. Please optimize
the concentration
based on your specific
assay requirements.

Immunogen Information

Gene ID

5970

Swiss Prot

Q04206

Immunogen

Synthetic peptide. This information is considered to be commercially sensitive.

Synonyms

p65; CMCU; NFKB3; AIF3BL3; IA

Product Information

Source

Rabbit

Isotype

IgG

Purification

Affinity purification

Storage

Store at -20°C. Avoid freeze / thaw cycles.

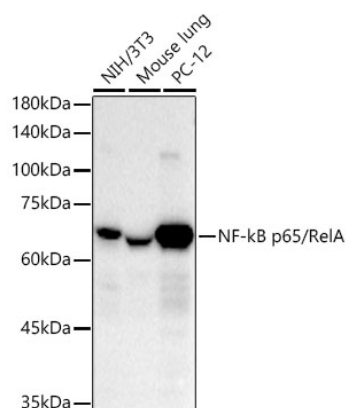
Buffer: PBS containing 50% glycerol and 0.05% BSA, preserved with proclin300 or sodium azide (as specified on the Certificate of Analysis), pH 7.3.

Contact

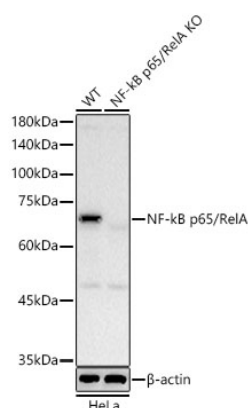


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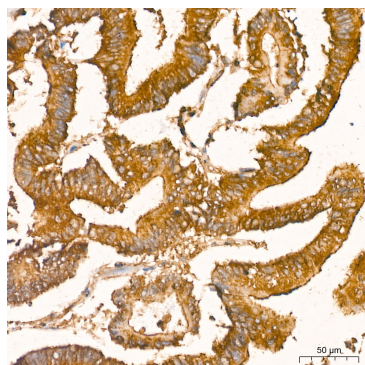
Validation Data



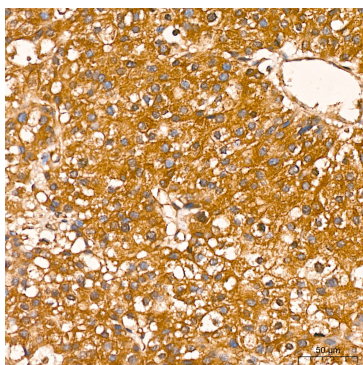
Western blot analysis of various lysates using [KO Validated] NF-kB p65/RelA Rabbit mAb (A22331) at 1:10000 dilution.
 Secondary antibody: HRP-conjugated Goat anti-Rabbit IgG (H+L) (AS014) at 1:10000 dilution.
 Lysates/proteins: 25µg per lane.
 Blocking buffer: 3% nonfat dry milk in TBST.
 Detection: ECL Basic Kit (RM00020).
 Exposure time: 30s.



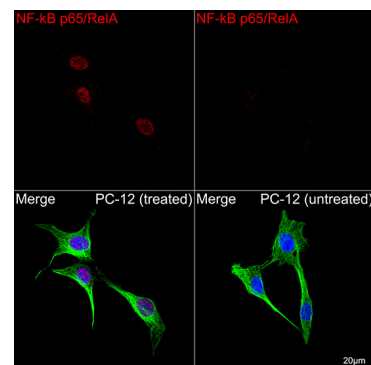
Western blot analysis of lysates from wild type (WT) and NF-kB p65/RelA knockout (KO) HeLa cells, using [KO Validated] NF-kB p65/RelA Rabbit mAb (A22331) at 1:10000 dilution.
 Secondary antibody: HRP-conjugated Goat anti-Rabbit IgG (H+L) (AS014) at 1:10000 dilution.
 Lysates/proteins: 25µg per lane.
 Blocking buffer: 3% nonfat dry milk in TBST.
 Detection: ECL Basic Kit (RM00020).
 Exposure time: 10s.



Immunohistochemistry analysis of paraffin-embedded Human colon carcinoma tissue using [KO Validated] NF-kB p65/RelA Rabbit mAb (A22331) at dilution of 1:200 (40x lens). High pressure antigen retrieval performed with 0.01M Citrate Buffer (pH 6.0) prior to IHC staining.

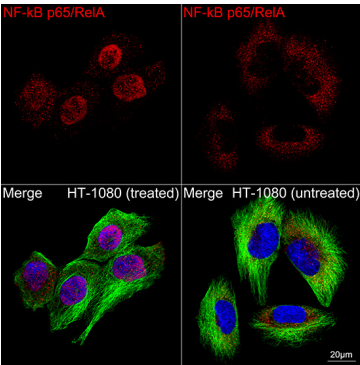


Immunohistochemistry analysis of paraffin-embedded Human liver tissue using [KO Validated] NF-kB p65/RelA Rabbit mAb (A22331) at dilution of 1:200 (40x lens). High pressure antigen retrieval performed with 0.01M Citrate Buffer (pH 6.0) prior to IHC staining.

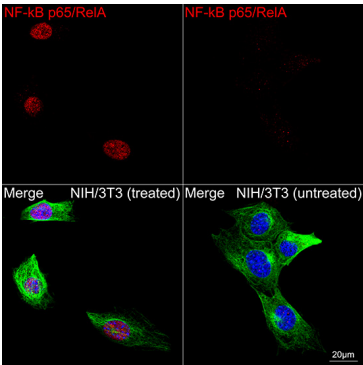


Confocal imaging of PC-12 cells (treated with TNF-α) and PC-12 cells (untreated) cells using [KO Validated] NF-kB p65/RelA Rabbit mAb (A22331, dilution 1:2000) followed by a further incubation with Cy3 Goat Anti-Rabbit IgG (H+L) (AS007, dilution 1:500) (Red). The cells were counterstained with α-Tubulin Mouse mAb (AC012, dilution 1:400) followed by incubation with ABflo® 488-conjugated Goat Anti-Mouse IgG (H+L) Ab (AS076, dilution 1:500) (Green). DAPI was used for nuclear staining (Blue). Objective: 100x.

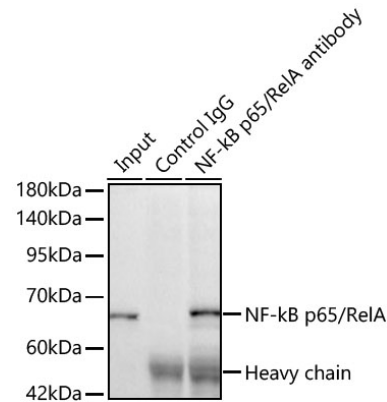
Validation Data



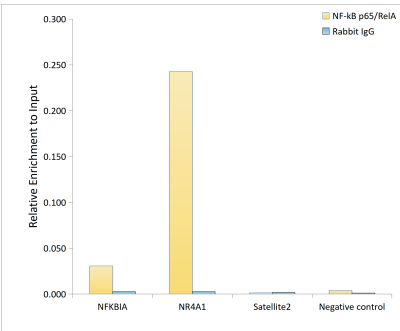
Confocal imaging of HT-1080 cells (treated with TNF-α) and HT-1080 cells (untreated) cells using [KO Validated] NF-kB p65/RelA Rabbit mAb (A22331, dilution 1:2000) followed by a further incubation with Cy3 Goat Anti-Rabbit IgG (H+L) (AS007, dilution 1:500) (Red). The cells were counterstained with α-Tubulin Mouse mAb (AC012, dilution 1:400) followed by incubation with ABflo® 488-conjugated Goat Anti-Mouse IgG (H+L) Ab (AS076, dilution 1:500) (Green). DAPI was used for nuclear staining (Blue). Objective: 100x.



Confocal imaging of NIH/3T3 cells (treated with TNF-α) and NIH/3T3 cells (untreated) cells using [KO Validated] NF-kB p65/RelA Rabbit mAb (A22331, dilution 1:2000) followed by a further incubation with Cy3 Goat Anti-Rabbit IgG (H+L) (AS007, dilution 1:500) (Red). The cells were counterstained with α-Tubulin Mouse mAb (AC012, dilution 1:400) followed by incubation with ABflo® 488-conjugated Goat Anti-Mouse IgG (H+L) Ab (AS076, dilution 1:500) (Green). DAPI was used for nuclear staining (Blue). Objective: 100x.



Immunoprecipitation of [KO Validated] NF-kB p65/RelA Rabbit mAb from 500 µg extracts of HeLa cells was performed using 2 µg of [KO Validated] NF-kB p65/RelA Rabbit mAb (A22331). Rabbit IgG isotype control (AC042) was used to precipitate the Control IgG sample. IP samples were eluted with 1X Laemmli Buffer. The Input lane represents 10% of the total input. Western blot analysis of immunoprecipitates was conducted using [KO Validated] NF-kB p65/RelA Rabbit mAb (A22331) at a dilution of 1:10000.



Chromatin immunoprecipitation analysis of extracts of HT-1080 cells, HT-1080 cells were treated by TNF-α (20 ng/ml) at 37°C for 30 minutes, using [KO Validated] NF-kB p65/RelA Rabbit mAb (A22331) and rabbit IgG. The amount of immunoprecipitated DNA was checked by quantitative PCR. Histogram was constructed by the ratios of the immunoprecipitated DNA to the input.