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

Catalog No.: A19653 KO Validated Recombinant 114 Publications

Basic Information

Observed MW

65kDa

Calculated MW

60kDa

Category

SMab Recombinant Monoclonal Antibody

Applications

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

Cross-Reactivity

Human, Mouse, Rat, Monkey

CloneNo number

ARC51086

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	1:2000 - 1:8000
IF/ICC	1:600 - 1:2400
ChIP	5μg antibody for 10μg-15μg of

ELISA Recommended starting concentration is 1

μg/mL. Please optimize the concentration based on your specific assay requirements.

Chromatin

Contact

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Immunogen Information

Gene ID	Swiss Prot
5970	Q04206

Immunogen

A synthetic peptide corresponding to a sequence within amino acids 450-551 of human NF-κB p65 (Q04206).

Synonyms

p65; CMCU; NFKB3; AIF3BL3; NF-kB p65/RelA

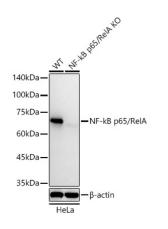
Product Information

Source	Isotype	Purification
Rabbit	IgG	Affinity purification

Storage

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

Buffer: PBS with 0.09% Sodium azide, 0.05% BSA, 50% glycerol, pH7.3.



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 (A19653) at 1:10000 dilution incubated overnight at 4° C.

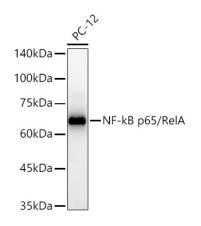
Secondary antibody: HRP-conjugated Goat anti-Rabbit IgG (H+L) (AS014) at 1:10000 dilution.

Lysates/proteins: 25 µg per lane.

Blocking buffer: 3% nonfat dry milk in TBST.

Detection: ECL Basic Kit (RM00020).

Exposure time: 30s.



Western blot analysis of lysates from PC-12 cells using [KO Validated] NF-kB p65/RelA Rabbit mAb (A19653) at 1:10000 dilution incubated overnight at 4°C.

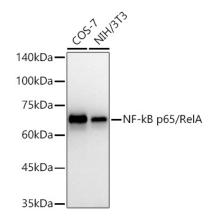
Secondary antibody: HRP-conjugated Goat anti-Rabbit IgG (H+L) (AS014) at 1:10000 dilution.

Lysates/proteins: 25 µg per lane.

Blocking buffer: 3% nonfat dry milk in TBST.

Detection: ECL Basic Kit (RM00020).

Exposure time: 30s.



Western blot analysis of various lysates using [KO Validated] NF-kB p65/RelA Rabbit mAb (A19653) at 1:10000 dilution incubated overnight at 4° C.

Secondary antibody: HRP-conjugated Goat anti-Rabbit IgG (H+L) (AS014) at 1:10000 dilution

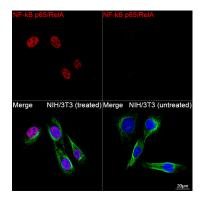
Lysates/proteins: 25 µg per lane.

Blocking buffer: 3% nonfat dry milk in TBST.

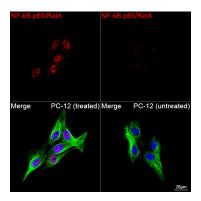
Detection: ECL Basic Kit (RM00020).

Exposure time: 60s.

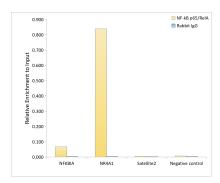
Validation Data



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 (A19653, dilution 1:2100) 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 PC-12 cells (treated with TNF- α) and PC-12 cells (untreated) cells using [KO Validated] NF-kB p65/RelA Rabbit mAb (A19653, dilution 1:2100) 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.



Chromatin immunoprecipitation was performed with 10 μ g of cross-linked chromatin from HT-1080 cells treated by TNF- α (20 ng/ml) at 37°C for 30 minutes, using 5 μ g of [KO Validated] NF-kB p65/RelA Rabbit mAb (A19653) and Rabbit IgG isotype control (AC042). The enrichment of immunoprecipitated DNA at different genomic loci was examined by quantitative PCR. The histogram compares the ratio of the immunoprecipitated DNA to the input at given loci.