

# [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

Primary antibody

### **Applications**

WB,IF/ICC,IP,ELISA,ChIP

### **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

**IF/ICC** 1:500 - 1:2000

**IP** 0.5μg-4μg antibody for

200μg-500μg extracts of

whole cells

**ChIP** 5μg antibody for

 $10\mu g$ - $15\mu g$  of Chromatin

**ELISA** Recommended starting

concentration is 1 µg/mL.

Please optimize the
concentration based on
your specific assay
requirements.

## **Contact**

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# Immunogen Information

**Gene ID**Swiss Prot
5970
Q04206

### **Immunogen**

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

### **Synonyms**

p65; CMCU; NFKB3; AIF3BL3; IA

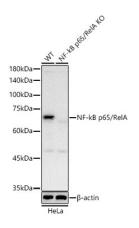
# **Product Information**

SourceIsotypePurificationRabbitIgGAffinity 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.



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.

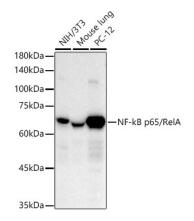
Secondary antibody: HRP-conjugated Goat anti-Rabbit IgG (H+L) (AS014) at 1:10000 dilution.

Lysates/proteins: 25µg per lane.

Blocking buffer: 3% nonfat dry milk in TBST.

Detection: ECL Basic Kit (RM00020).

Exposure time: 10s.



Western blot analysis of various lysates using [KO Validated] NF-kB p65/RelA Rabbit mAb (A22331) at 1:10000 dilution.

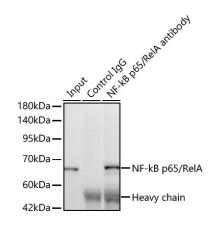
Secondary antibody: HRP-conjugated Goat anti-Rabbit IgG (H+L) (AS014) at 1:10000 dilution.

Lysates/proteins: 25µg per lane.

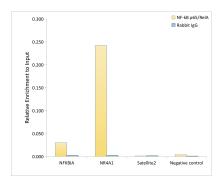
Blocking buffer: 3% nonfat dry milk in TBST.

Detection: ECL Basic Kit (RM00020).

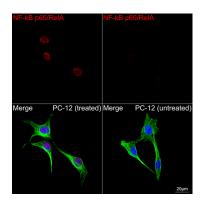
Exposure time: 30s.



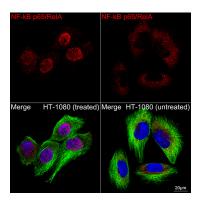
Immunoprecipitation of [KO Validated] NF-kB p65/RelA Rabbit mAb from 500  $\mu$ g extracts of HeLa cells was performed using 2  $\mu$ g of [KO Validated] NF-kB p65/RelA Rabbit mAb (A22331). Rabbit IgG isotype control (AC042) was used to precipitate the Control IgG sample. IP samples were eluted with 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.



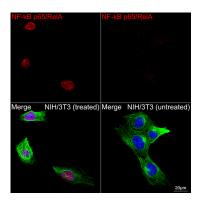
Chromatin immunoprecipitation analysis of extracts of HT-1080 cells, HT-1080 cells were treated by TNF- $\alpha$  (20 ng/ml) at 37°C for 30 minutes, using [KO Validated] NF-kB p65/RelA Rabbit mAb (A22331) and rabbit lgG.The amount of immunoprecipitated DNA was checked by quantitative PCR. Histogram was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



Confocal imaging of PC-12 cells (treated with TNF- $\alpha$ ) 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  $\alpha$ -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 HT-1080 cells (treated with TNF- $\alpha$ ) 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  $\alpha$ -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- $\alpha$ ) 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  $\alpha$ -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.