

Anti-NF- κ B p65 Antibody

ER0815



Product Type:	Rabbit polyclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat, Zebrafish
Applications:	WB, IHC-P, FC
Molecular Wt:	Predicted band size: 60 kDa

Description: NF-kappa-B is a pleiotropic transcription factor present in almost all cell types and is the endpoint of a series of signal transduction events that are initiated by a vast array of stimuli related to many biological processes such as inflammation, immunity, differentiation, cell growth, tumorigenesis and apoptosis. NF-kappa-B is a homo- or heterodimeric complex formed by the Rel-like domain-containing proteins RELA/p65, RELB, NFKB1/p105, NFKB1/p50, REL and NFKB2/p52 and the heterodimeric p65-p50 complex appears to be most abundant one. In unstimulated cells, NF- κ B is sequestered in the cytoplasm by I κ B inhibitory proteins. NF- κ B-activating agents can induce the phosphorylation of I κ B proteins, targeting them for rapid degradation through the ubiquitin-proteasome pathway and releasing NF- κ B to enter the nucleus where it regulates gene expression.

Immunogen: Synthetic peptide within N-terminal human NF- κ B p65.

Positive control: Hela cell lysate, A549 cell lysate, PC12 cell lysate, Mouse embryonic stem cell lysate, NIH/3T3 cell lysate, zebrafish lysates, human lung cancer tissue, human lung tissue, human spleen tissue, mouse spleen tissue, rat spleen tissue, zebrafish, Hela.

Subcellular location: Nucleus, cytoplasm.

Database links: SwissProt: Q04206 Human | Q04207 Mouse
Entrez Gene: 309165 Rat

Recommended Dilutions:

WB	1:1,000-1:2,000
IHC-P	1:200
FC	1:50-1:100

Storage Buffer: 1*PBS (pH7.4), 0.2% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

Storage Instruction: Store at +4°C after thawing. Aliquot store at -20°C. Avoid repeated freeze / thaw cycles.

Purity: Immunogen affinity purified.

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Images

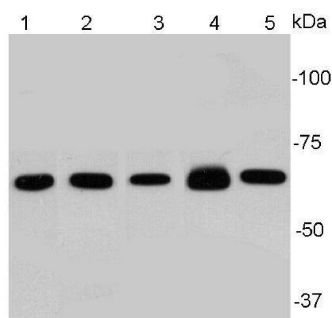


Fig1: Western blot analysis of NF-κB p65 on different lysates using anti-NF-κB p65 antibody at 1/1,000 dilution.

Positive control:

- Lane 1: Hela cell lysate
- Lane 2: A549 cell lysate
- Lane 3: PC12 cell lysate
- Lane 4: Mouse embryonic stem cell lysate
- Lane 5: NIH/3T3 cell lysate

Fig2: Western blot analysis of NF-κB p65 on zebrafish lysates with Rabbit anti-NF-κB p65 antibody (ER0815) at 1/2,000 dilution.

Lysates at 20 μg/Lane.

Predicted band size: 60kDa
Observed band size: 70 kDa

Exposure time: 3min20s;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDm/TBST for 1 hour at room temperature. The primary antibody (ER0815) at 1/2,000 dilution was used in 5% NFDm/TBST at 4°C overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:50,000 dilution was used for 1 hour at room temperature.

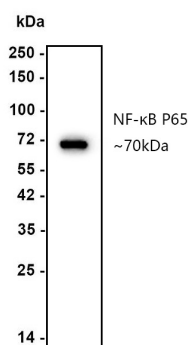
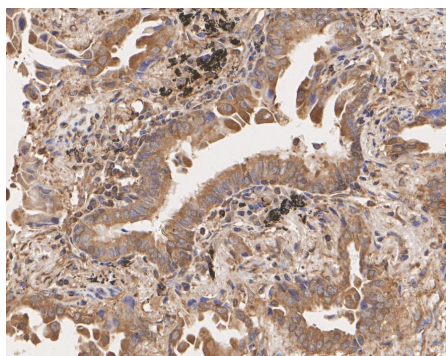


Fig3: Immunohistochemical analysis of paraffin-embedded human lung cancer tissue with Rabbit anti-NF-κB p65 antibody (ER0815) at 1/200 dilution.



The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ER0815) at 1/200 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

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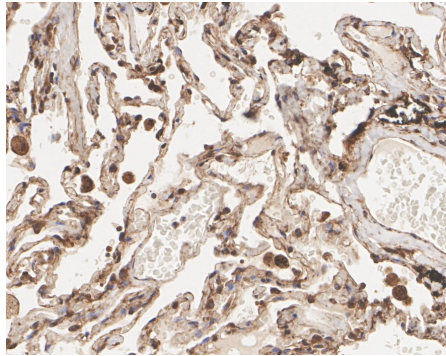


Fig4: Immunohistochemical analysis of paraffin-embedded human lung tissue with Rabbit anti-NF-κB p65 antibody (ER0815) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ER0815) at 1/200 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

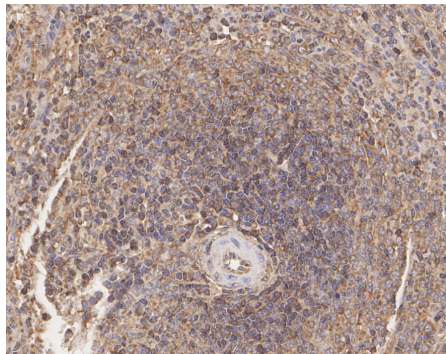


Fig5: Immunohistochemical analysis of paraffin-embedded human spleen tissue with Rabbit anti-NF-κB p65 antibody (ER0815) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ER0815) at 1/200 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

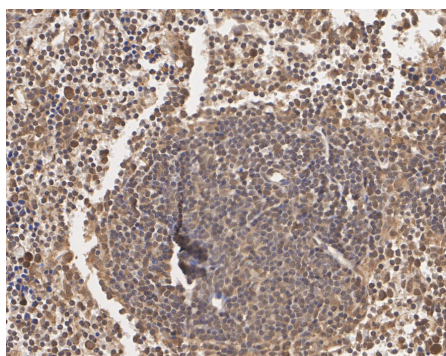


Fig6: Immunohistochemical analysis of paraffin-embedded mouse spleen tissue with Rabbit anti-NF-κB p65 antibody (ER0815) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ER0815) at 1/200 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

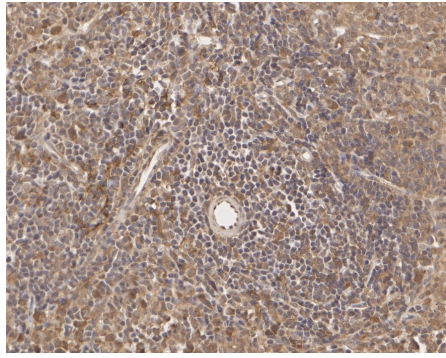


Fig7: Immunohistochemical analysis of paraffin-embedded rat spleen tissue with Rabbit anti-NF-κB p65 antibody (ER0815) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ER0815) at 1/200 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

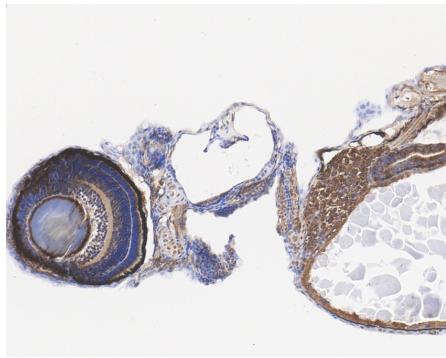


Fig8: Immunohistochemical analysis of paraffin-embedded zebrafish with Rabbit anti-NF-κB p65 antibody (ER0815) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ER0815) at 1/200 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

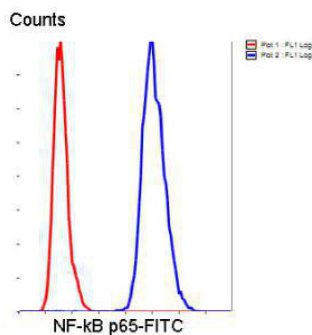


Fig9: Flow cytometric analysis of HeLa cells with NF-κB p65 antibody at 1/50 dilution (blue) compared with an unlabelled control (cells without incubation with primary antibody; red). Goat anti rabbit IgG (FITC) was used as the secondary antibody.

Note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

Background References

1. "Duration of nuclear NF-kappaB action regulated by reversible acetylation." Chen L.F., Fischle W., Verdin E., Greene W.C. *Science* 293:1653-1657(2001)
2. "A novel protein overexpressed in hepatoma accelerates export of NF-kappa B from the nucleus and inhibits p53-dependent apoptosis." Higashitsuji H., Higashitsuji H., Nagao T., Nonoguchi K., Fujii S., Itoh K., Fujita J. *Cancer Cell* 2:335-346(2002)
3. "Breast cancer metastasis suppressor 1 functions as a corepressor by enhancing histone deacetylase 1-mediated deacetylation of RelA/p65 and promoting apoptosis." Liu Y., Smith P.W., Jones D.R. *Mol. Cell. Biol.* 26:8683-8696(2006)
4. "SIRT2 regulates NF-kappaB dependent gene expression through deacetylation of p65 Lys310." Rothgiesser K.M., Erener S., Waibel S., Luscher B., Hottiger M.O. *J. Cell Sci.* 123:4251-4258(2010)

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