

Product datasheet

Anti-NFkB p105 / p50 antibody [E381] ab32360

敲除验证
重组
RabMAb

★★★★★
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概述

产品名称	Anti-NFkB p105 / p50抗体[E381]
描述	兔单克隆抗体[E381] to NFkB p105 / p50
宿主	Rabbit
特异性	This antibody will detect both forms: p50 and p105.
经测试应用	适用于: WB, IHC-P 不适用于: Flow Cyt
种属反应性	与反应: Mouse, Rat, Human
免疫原	Synthetic peptide (the amino acid sequence is considered to be commercially sensitive) corresponding to NFkB p105/ p50 aa 300-400.
阳性对照	WB: HeLa, MCF-7 and PC-12 cell lysates. IHC-P: human prostate carcinoma and bladder carcinoma tissues.
常规说明	<p>Our RabMAb[®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMab[®] patents</p> <p>We are constantly working hard to ensure we provide our customers with best in class antibodies. As a result of this work we are pleased to now offer this antibody in purified format. We are in the process of updating our datasheets. The purified format is designated 'PUR' on our product labels. If you have any questions regarding this update, please contact our Scientific Support team.</p> <p>This product is a recombinant rabbit monoclonal antibody.</p>

性能

形式	Liquid
存放说明	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C. Avoid freeze / thaw cycle.
存储溶液	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol, 0.05% BSA
纯度	Protein A purified

克隆	单克隆
克隆编号	E381
同种型	IgG

应用

Our [Abpromise guarantee](#) covers the use of **ab32360** in the following tested applications.

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

应用	Ab评论	说明
WB	★★★★★	1/1000 - 1/10000. Detects a band of approximately 50, 105 kDa (predicted molecular weight: 50 kDa).
IHC-P	★★★★☆	1/250 - 1/500. See protocols (link: http://www.abcam.com/protocols/ihc-antigen-retrieval-protocol).

应用说明 Is unsuitable for Flow Cyt.

靶标

功能 NF-kappa-B is a pleiotropic transcription factor which is present in almost all cell types and is involved in 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. The dimers bind at kappa-B sites in the DNA of their target genes and the individual dimers have distinct preferences for different kappa-B sites that they can bind with distinguishable affinity and specificity. Different dimer combinations act as transcriptional activators or repressors, respectively. NF-kappa-B is controlled by various mechanisms of post-translational modification and subcellular compartmentalization as well as by interactions with other cofactors or corepressors. NF-kappa-B complexes are held in the cytoplasm in an inactive state complexed with members of the NF-kappa-B inhibitor (I-kappa-B) family. In a conventional activation pathway, I-kappa-B is phosphorylated by I-kappa-B kinases (IKKs) in response to different activators, subsequently degraded thus liberating the active NF-kappa-B complex which translocates to the nucleus. NF-kappa-B heterodimeric p65-p50 and RelB-p50 complexes are transcriptional activators. The NF-kappa-B p50-p50 homodimer is a transcriptional repressor, but can act as a transcriptional activator when associated with BCL3. NFKB1 appears to have dual functions such as cytoplasmic retention of attached NF-kappa-B proteins by p105 and generation of p50 by a cotranslational processing. The proteasome-mediated process ensures the production of both p50 and p105 and preserves their independent function, although processing of NFKB1/p105 also appears to occur post-translationally. p50 binds to the kappa-B consensus sequence 5'-GGRNNYCC-3', located in the enhancer region of genes involved in immune response and acute phase reactions. In a complex with MAP3K8, NFKB1/p105 represses MAP3K8-induced MAPK signaling; active MAP3K8 is released by proteasome-dependent degradation of NFKB1/p105.

序列相似性 Contains 7 ANK repeats.
Contains 1 death domain.
Contains 1 RHD (Rel-like) domain.

结构域

The C-terminus of p105 might be involved in cytoplasmic retention, inhibition of DNA-binding, and transcription activation.

Glycine-rich region (GRR) appears to be a critical element in the generation of p50.

翻译后修饰

While translation occurs, the particular unfolded structure after the GRR repeat promotes the generation of p50 making it an acceptable substrate for the proteasome. This process is known as cotranslational processing. The processed form is active and the unprocessed form acts as an inhibitor (I kappa B-like), being able to form cytosolic complexes with NF-kappa B, trapping it in the cytoplasm. Complete folding of the region downstream of the GRR repeat precludes processing.

Phosphorylation at 'Ser-903' and 'Ser-907' primes p105 for proteolytic processing in response to TNF-alpha stimulation. Phosphorylation at 'Ser-927' and 'Ser-932' are required for BTRC/BTRCP-mediated proteolysis.

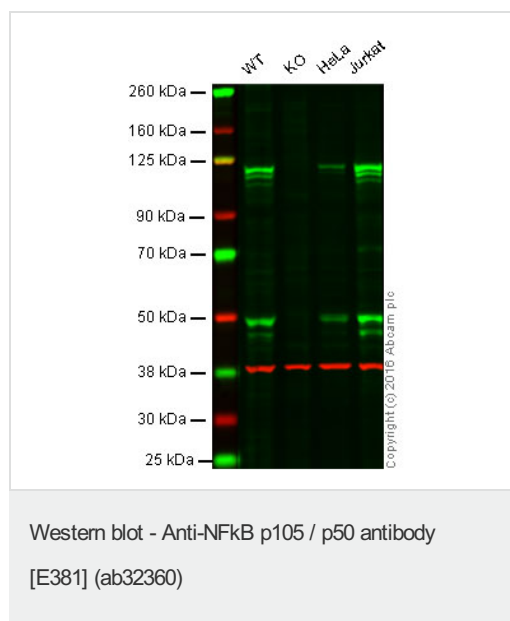
Polyubiquitination seems to allow p105 processing.

S-nitrosylation of Cys-61 affects DNA binding.

细胞定位

Nucleus. Cytoplasm. Nuclear, but also found in the cytoplasm in an inactive form complexed to an inhibitor.

图片



Lane 1: Wild-type HAP1 cell lysate (20 μg)

Lane 2: NFκB p105 / p50 knockout HAP1 cell lysate (20 μg)

Lane 3: HeLa cell lysate (20 μg)

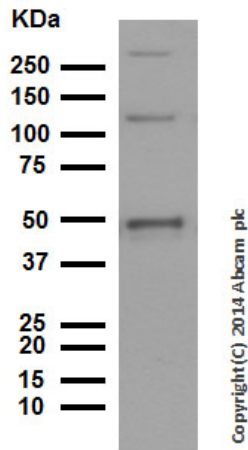
Lane 4: Jurkat cell lysate (20 μg)

Lanes 1 - 4: Merged signal (red and green).

Green - ab32360 observed at 120, 50 kDa.

Red - loading control, ab8245, observed at 37 kDa.

ab32360 was shown to specifically react with NFκB p105 / p50 when NFκB p105 / p50 knockout samples were used. Wild-type and NFκB p105 / p50 knockout samples were subjected to SDS-PAGE. ab32360 and ab8245 (loading control to GAPDH) were diluted at 1/1000 and 1/10 000 respectively and incubated overnight at 4°C. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (ab216776) secondary antibodies at 1/10000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-NFkB p105 / p50 antibody [E381] (ab32360)

Anti-NFkB p105 / p50 antibody [E381] (ab32360) at 1/10000 dilution (purified) + HeLa cell lysate at 20 µg

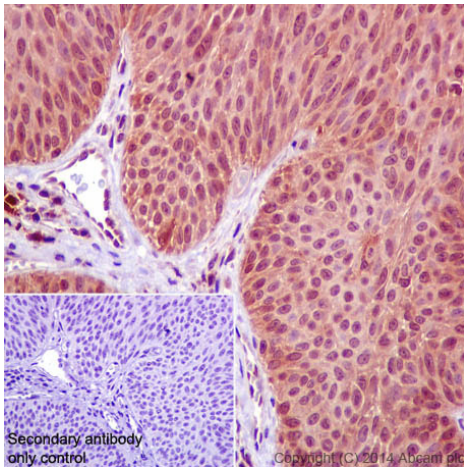
Secondary

Peroxidase conjugated goat anti-rabbit IgG (H+L) at 1/1000 dilution

Predicted band size: 50 kDa

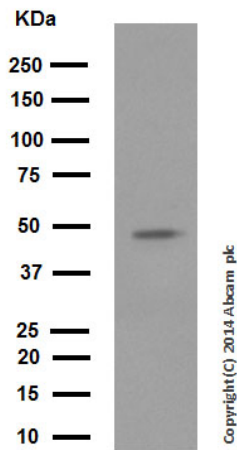
Observed band size: 105,50 kDa

Blocking and dilution buffer: 5% NFDm/TBST.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NFkB p105 / p50 antibody [E381] (ab32360)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human bladder carcinoma tissue labelling NFkB p105 / p50 with purified ab32360 at 1/250. Heat mediated antigen retrieval was performed using Tris/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.



Western blot - Anti-NFkB p105 / p50 antibody [E381] (ab32360)

Anti-NFkB p105 / p50 antibody [E381] (ab32360) at 1/50000 dilution (purified) + MCF-7 cell lysate at 10 µg

Secondary

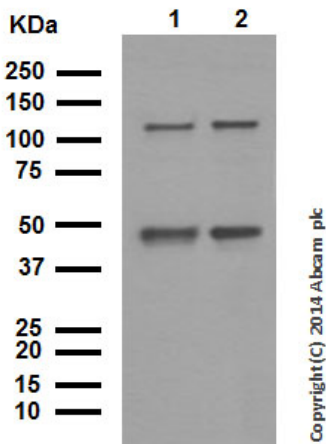
Peroxidase conjugated goat anti-rabbit IgG (H+L) at 1/1000 dilution

Predicted band size: 50 kDa

Observed band size: 105 kDa

Additional bands at: 50 kDa (possible non-specific binding)

Blocking and dilution buffer: 5% NFDm/TBST.



Western blot - Anti-NFkB p105 / p50 antibody [E381] (ab32360)

All lanes : Anti-NFkB p105 / p50 antibody [E381] (ab32360) at 1/10000 dilution (purified)

Lane 1 : PC-12 cell lysate

Lane 2 : NIH/3T3 cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

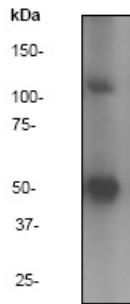
All lanes : Peroxidase conjugated goat anti-rabbit IgG (H+L) at 1/1000 dilution

Predicted band size: 50 kDa

Observed band size: 105 kDa

Additional bands at: 50 kDa (possible non-specific binding)

Blocking and dilution buffer: 5% NFDm/TBST.

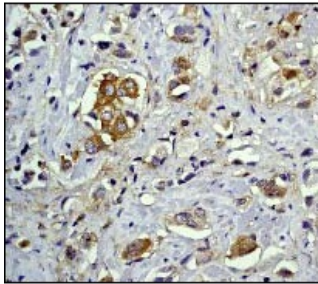


Anti-NFκB p105 / p50 antibody [E381]
(ab32360) at 1/5000 dilution (unpurified) +
HeLa cell lysate

Predicted band size: 50 kDa

Observed band size: 105,50 kDa

Western blot - Anti-NFκB p105 / p50 antibody
[E381] (ab32360)



Immunohistochemistry (Formalin/PFA-fixed
paraffin-embedded sections) analysis of
human prostate carcinoma tissue labelling
NFκB p105 / p50 with unpurified ab32360 at a
dilution of 1/250.

Immunohistochemistry (Formalin/PFA-fixed paraffin-
embedded sections) - Anti-NFκB p105 / p50
antibody [E381] (ab32360)

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