abcam

Product datasheet

Anti-p95/NBS1 antibody [Y112] ab32074

敲除验证 重组 RabMAb

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概述

产品名称	Anti-p95/NBS1抗体[Y112]
描述	兔单克隆抗体[Y112] to p95/NBS1
宿主	Rabbit
经测试应用	**适用于:** WB, IHC-P **不适用于:** Flow Cyt or ICC/IF
种属反应性	**与反应:** Human **预测可用于:** Mouse, Rat
免疫原	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
阳性对照	WB: HeLa, Jurkat and Wild-type A431 cell lysates. IHC-P: Human testis and skin carcinoma tissues.
常规说明	This product is a recombinant monoclonal antibody, which offers several advantages including: - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production For more information **see here**. Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb® patents**.

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 (glycerin, glycerine), 0.05% BSA
纯度	Protein A purified
克隆	单克隆

克隆编号	Y112
同种型	IgG

应用

The Abpromise guarantee **Abpromise™承诺保证使用ab32074于以下的经测试应用**

"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
WB	★★★★☆ (3)	1/1000 - 1/10000. Predicted molecular weight: 85 kDa. **For unpurified use at 1/1000 - 1/2000.**
IHC-P	★★★★★ (1)	1/200. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. See **IHC antigen retrieval protocols**. **For unpurified use at 1/50.**

应用说明 Is unsuitable for Flow Cyt or ICC/IF.

靶标

功能 Component of the MRE11-RAD50-NBN (MRN complex) which plays a critical role in the cellular response to DNA damage and the maintenance of chromosome integrity. The complex is involved in double-strand break (DSB) repair, DNA recombination, maintenance of telomere integrity, cell cycle checkpoint control and meiosis. The complex possesses single-strand endonuclease activity and double-strand-specific 3'-5' exonuclease activity, which are provided by MRE11A. RAD50 may be required to bind DNA ends and hold them in close proximity. NBN modulate the DNA damage signal sensing by recruiting PI3/PI4-kinase family members ATM, ATR, and probably DNA-PKcs to the DNA damage sites and activating their functions. It can also recruit MRE11 and RAD50 to the proximity of DSBs by an interaction with the histone H2AX. NBN also functions in telomere length maintenance by generating the 3' overhang which serves as a primer for telomerase dependent telomere elongation. NBN is a major player in the control of intra-S-phase checkpoint and there is some evidence that NBN is involved in G1 and G2 checkpoints. The roles of NBS1/MRN encompass DNA damage sensor, signal transducer, and effector, which enable cells to maintain DNA integrity and genomic stability. Forms a complex with RBBP8 to link DNA double-strand break sensing to resection. Enhances AKT1 phosphorylation possibly by association with the mTORC2 complex.

组织特异性 Ubiquitous. Expressed at high levels in testis.

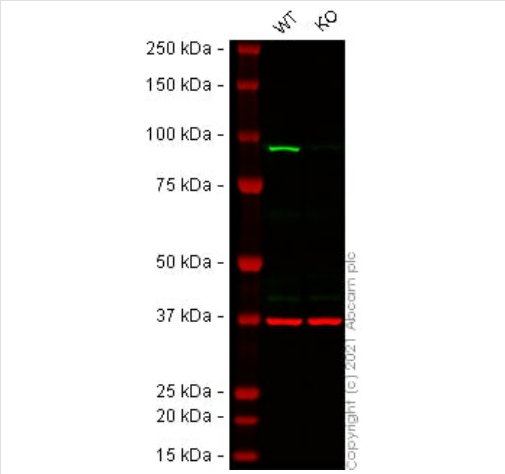
疾病相关 Nijmegen breakage syndrome
Breast cancer
Aplastic anemia
Defects in NBN might play a role in the pathogenesis of childhood acute lymphoblastic leukemia (ALL).

序列相似性 Contains 1 BRCT domain.
Contains 1 FHA domain.

结构域 The FHA and BRCT domains are likely to have a crucial role for both binding to histone H2AFX and for relocalization of MRE11/RAD50 complex to the vicinity of DNA damage.

	The C-terminal domain contains a MRE11-binding site, and this interaction is required for the nuclear localization of the MRN complex. The EEXXXDDL motif at the C-terminus is required for the interaction with ATM and its recruitment to sites of DNA damage and promote the phosphorylation of ATM substrates, leading to the events of DNA damage response.
翻译后修饰	Phosphorylated by ATM in response of ionizing radiation, and such phosphorylation is responsible intra-S phase checkpoint control and telomere maintenance.
细胞定位	Nucleus. Nucleus, PML body. Chromosome, telomere. Localizes to discrete nuclear foci after treatment with genotoxic agents.

图片



Western blot - Anti-p95/NBS1 antibody [Y112] (ab32074)

All lanes : Anti-p95/NBS1 antibody [Y112] (ab32074) at 1/1000 dilution

Lane 1 : Wild-type A431 cell lysate
Lane 2 : NBN knockout A431 cell lysate

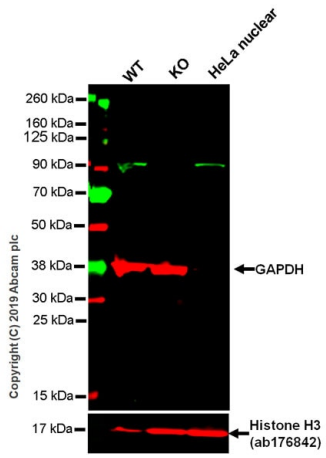
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 85 kDa
Observed band size: 90 kDa

Lanes 1 - 2: Merged signal (red and green). Green - ab32074 observed at 90 kDa. Red - loading control **ab8245** (Mouse anti-GAPDH antibody [6C5]) observed at 37 kDa.

ab32074 was shown to react with p95/NBS1 in wild-type A431 cells in Western blot with loss of signal observed in NBN knockout cell line **ab269506** (NBN knockout cell lysate **ab269668**). Wild-type A431 and NBN knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 5 % milk in TBS-T (0.1 % Tween®) before incubation with ab32074 and **ab8245** (Mouse anti-GAPDH antibody [6C5]) overnight at 4 °C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 h at room temperature before imaging.



Western blot - Anti-p95/NBS1 antibody [Y112] (ab32074)

All lanes : Anti-p95/NBS1 antibody [Y112] (ab32074) at 1/1000 dilution

Lane 1 : Wild-type HeLa cell lysate
Lane 2 : NBN knockout HeLa cell lysate
Lane 3 : Wild-type HeLa nuclear cell lysate

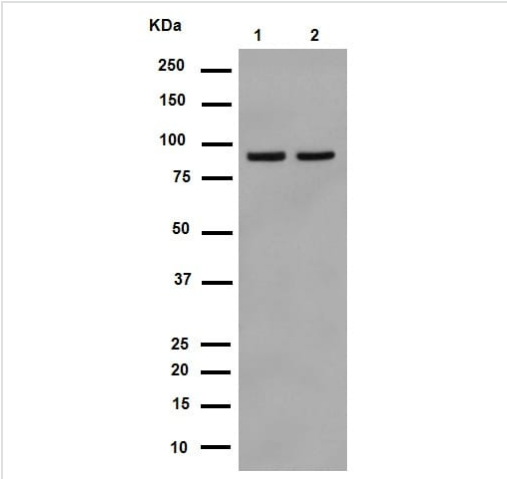
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 85 kDa
Observed band size: 95 kDa

Lanes 1-3: Merged signal (red and green). Green - ab32074 observed at 95 kDa.

ab32074 Anti-p95/NBS1 antibody [Y112] was shown to specifically react with p95/NBS1 in wild-type HeLa cells. Loss of signal was observed when knockout cell line **ab261834** (knockout cell lysate **ab257111**) was used. Wild-type and p95/NBS1 knockout samples were subjected to SDS-PAGE. ab32074, Anti-GAPDH antibody [6C5] - Cytoplasmic Loading Control (**ab8245**) and Anti-Histone H3 (**ab176842**) - Nuclear Loading Control were incubated overnight at 4°C at 1 in 1000 dilution, 1 in 20000 dilution and 1 in 1000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (**ab216773**), Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (**ab216776**) secondary antibodies and Goat anti-Rabbit IgG H&L (IRDye® 680RD) preadsorbed (**ab216777**) at 1 in 10000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-p95/NBS1 antibody [Y112] (ab32074)

All lanes : Anti-p95/NBS1 antibody [Y112] (ab32074) at 1/1200 dilution (unpurified)

Lane 1 : HeLa cell lysate
Lane 2 : Jurkat cell lysate

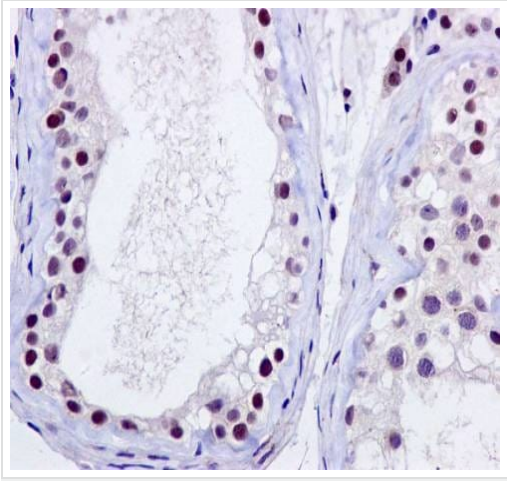
Lysates/proteins at 20 µg per lane.

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

Predicted band size: 85 kDa
Observed band size: 95 kDa

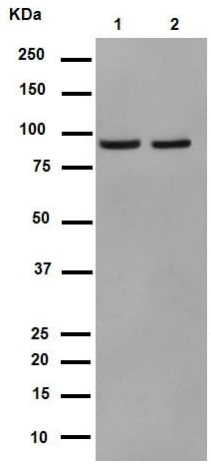
Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-p95/NBS1 antibody [Y112] (ab32074)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human testis tissue labelling p95/NBS1 with purified ab32074 at 1/200. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. A prediluted HRP-polymer conjugated anti-rabbit IgG was used as the secondary antibody. Counterstained with hematoxylin.



Western blot - Anti-p95/NBS1 antibody [Y112] (ab32074)

All lanes : Anti-p95/NBS1 antibody [Y112] (ab32074) at 1/10000 dilution (purified)

Lane 1 : HeLa cell lysate
Lane 2 : Jurkat cell lysate

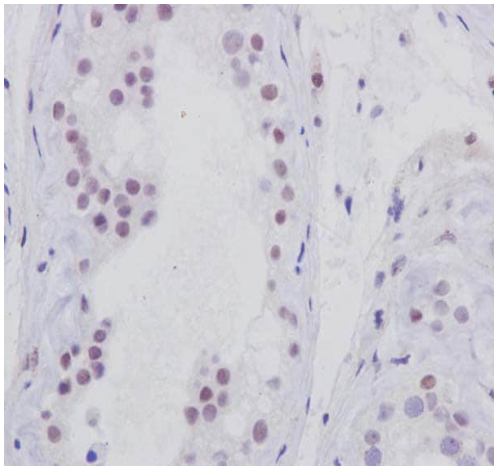
Lysates/proteins at 20 µg per lane.

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

Predicted band size: 85 kDa
Observed band size: 95 kDa

Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-p95/NBS1 antibody [Y112] (ab32074)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human testis tissue labelling p95/NBS1 with unpurified ab32074 at 1/20. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. A prediluted HRP-polymer conjugated anti-rabbit IgG was used as the secondary antibody. Counterstained with hematoxylin.

Anti-p95/NBS1 antibody [Y112] (ab32074)

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