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Product datasheet

Anti-BNIP3 antibody [ANa40] ab10433

敲除验证

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概述

产品名称	Anti-BNIP3抗体[ANa40]
描述	小鼠单克隆抗体[ANa40] to BNIP3
宿主	Mouse
经测试应用	**适用于:** WB, IHC-P, Flow Cyt (Intra)
种属反应性	**与反应:** Human **预测可用于:** Mouse, Rat
免疫原	Recombinant fragment corresponding to Human BNIP3.
表位	The epitope recognized by the antibody resides within amino acids 112-124 of human BNIP3 molecule.
阳性对照	This antibody gave a positive signal in human skeletal muscle tissue lysate in western blot and on human kidney formalin-fixed, paraffin-embedded tissue sections in IHC.
常规说明	This antibody clone is manufactured by Abcam. If you require a custom buffer formulation or conjugation for your experiments, please contact **orders@abcam.com**. The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets your needs before purchasing. If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, along with publications, customer reviews and Q&As

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 or -80°C. Avoid freeze / thaw cycle.
存储溶液	pH: 7.40 Preservative: 0.02% Sodium azide Constituents: PBS, 6.97% L-Arginine

纯度	Protein G purified
克隆	单克隆
克隆编号	ANa40
骨髓瘤	unknown
同种型	IgG2b
轻链类型	kappa

应用

The Abpromise guarantee **Abpromise™承诺保证使用ab10433于以下的经测试应用**

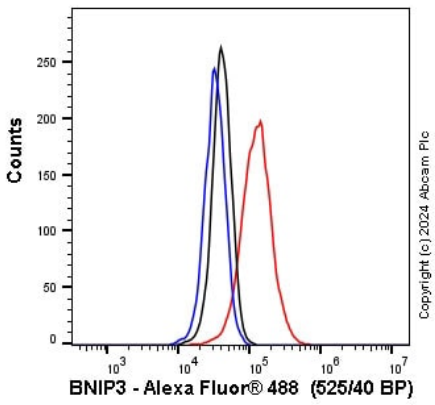
"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
WB	★★★★☆ (5)	Use a concentration of 1 - 5 µg/ml. Detects a band of approximately 30 kDa (predicted molecular weight: 30 kDa). We recommend using 3% milk as the blocking agent for Western blot.
IHC-P		Use a concentration of 5 µg/ml. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
Flow Cyt (Intra)		Use 0.1-1µg for 10^6 cells. **ab170192** - Mouse monoclonal IgG2b, is suitable for use as an isotype control with this antibody.

靶标

功能	Apoptosis-inducing protein that, which can overcome BCL2 suppression. May play a role in repartitioning calcium between the two major intracellular calcium stores in association with BCL2.
序列相似性	Belongs to the NIP3 family.
细胞定位	Mitochondrion. Mitochondrion membrane. Coexpression with the EIB 19-kDa protein results in a shift in NIP3 localization pattern to the nuclear envelope. Colocalizes with ACAA2 in the mitochondria.

图片



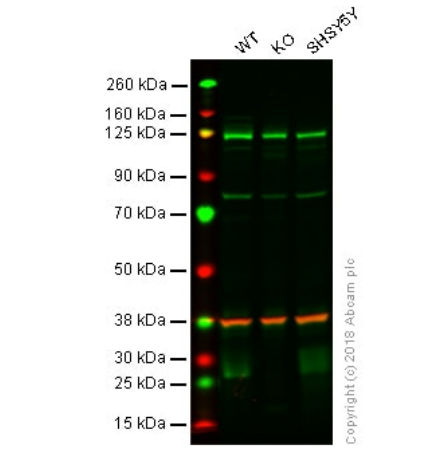
Flow Cytometry (Intracellular) - Anti-BNIP3 antibody [ANa40] (ab10433)

Flow cytometry overlay histogram showing HepG2 cells stained with ab10433 (red line). The cells were fixed with 4% paraformaldehyde (10 min) and then permeabilized with 0.1% PBS-Triton X-100 for 15 min. The cells were then incubated in 1x PBS containing 10% normal goat serum to block non-specific protein-protein interaction followed by the antibody (ab10433) ($1x10^6$ in 100µl at 5.0µg/ml) for 30 min at 22°C.

The secondary antibody Goat Anti-Mouse IgG H&L (Alexa Fluor® 488) (**ab150117**) preadsorbed was incubated at 1/4000 for 30 min at 22°C.

Isotype control antibody (black line) was Mouse IgG2b, kappa monoclonal [7E10G10] (**ab170192**) used under the same concentration and conditions as the primary antibody. Unlabelled sample (blue line) was also used as a control.

Acquisition of >5000 events were collected using a 50 mW Blue laser (488nm) and 525/40 bandpass filter.

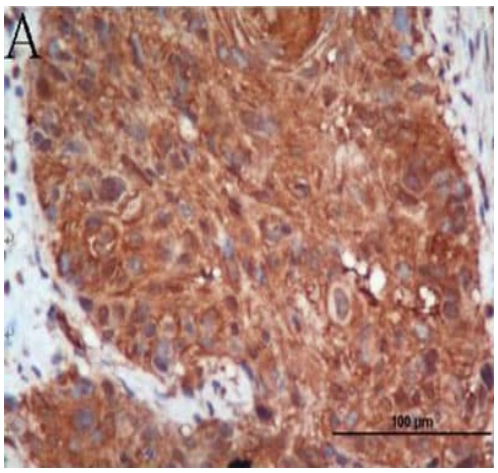


Western blot - Anti-BNIP3 antibody [ANa40] (ab10433)

Lane 1: Wild-type HAP1 whole cell lysate (40 µg)
Lane 2: BNIP3 knockout HAP1 whole cell lysate (40 µg)
Lane 3: SHSY5Y whole cell lysate (40 µg)

Lanes 1 - 3: Merged signal (red and green). Green - ab10433 observed at 30 kDa. Red - loading control, **ab181602**, observed at 37 kDa.

ab10433 was shown to recognize BNIP3 in wild-type HAP1 cells as signal was lost at the expected MW in BNIP3 knockout cells. Additional cross-reactive bands were observed in the wild-type and knockout cells. Wild-type and BNIP3 knockout samples were subjected to SDS-PAGE. ab10433 and **ab181602** (Rabbit anti-GAPDH loading control) were incubated overnight at 4°C at 5 µg/mL and 1/20000 dilution respectively. Blots were developed with Goat anti-Mouse IgG H&L (IRDye® 800CW) preabsorbed **ab216772** and Goat anti-Rabbit IgG H&L (IRDye® 680RD) preabsorbed **ab216777** secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.

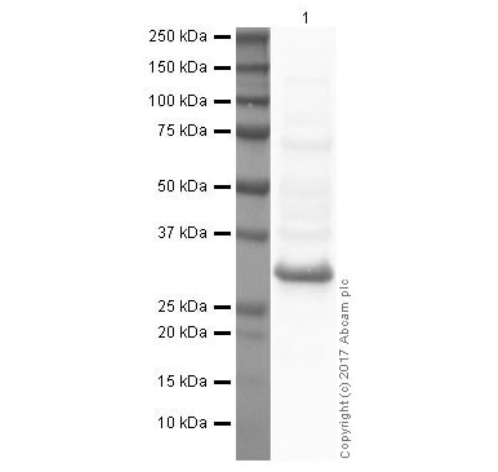


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-BNIP3 antibody [ANa40] (ab10433)

Image from Jin T et al., J Transl Med. 2012 Mar 30;10:64 Fig 1.; doi:10.1186/1479-5876-10-64; 30 March 2012, Journal of Translational Medicine 2012, 10:64

Immunohistochemical analysis of Human laryngeal squamous cell carcinoma tissue, staining BNIP3 with ab10433.

Antigen retrieval was performed by heat mediation in citrate buffer. The sections were incubated with primary antibody (1/100) overnight at 4°C in a humidified chamber. Staining was visualized using DAB, followed by hematoxylin nuclear counterstaining.



Western blot - Anti-BNIP3 antibody [ANa40] (ab10433)

Anti-BNIP3 antibody [ANa40] (ab10433) at 1 µg/ml + Human skeletal muscle tissue lysate - total protein (**ab29330**) at 20 µg

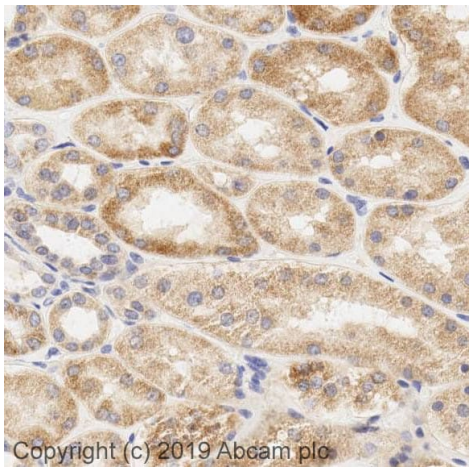
Secondary
Goat polyclonal to Mouse IgG - H&L - Pre-Adsorbed (HRP) (**ab65485**) at 1/5000 dilution

Performed under reducing conditions.

Predicted band size: 30 kDa

Exposure time: 5 seconds

This blot was produced using a 4-12% Bis-tris gel under the MOPS buffer system. The gel was run at 200V for 50 minutes before being transferred onto a Nitrocellulose membrane at 30V for 70 minutes. The membrane was then blocked for an hour using 3% milk before being incubated with ab10433 overnight at 4°C. Antibody binding was detected using an anti-mouse antibody conjugated to HRP, and visualised using ECL development solution **ab133406**.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-BNIP3 antibody [ANa40] (ab10433)

IHC image of BNIP3 antibody staining in a section of formalin-fixed paraffin-embedded normal human kidney* performed on a Leica BOND™ system using the standard protocol. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20mins. The section was then incubated with ab10433, 1ug/ml, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX.

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.

**Tissue obtained from the Human Research Tissue Bank, supported by the NIHR Cambridge Biomedical Research Centre*

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