

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human BNIP3 in direct ELISAs.
Source	Monoclonal Mouse IgG _{2A} Clone # 670639
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	<i>E. coli</i> -derived recombinant human BNIP3 Gly2-Ser160 Accession # Q12983
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied either lyophilized or as a 0.2 µm filtered solution in PBS.

APPLICATIONS

Please Note: Optimal dilutions should be determined by each laboratory for each application. *General Protocols* are available in the *Technical Information* section on our website.

	Recommended Concentration	Sample
Western Blot	0.5 µg/mL	See Below
Immunocytochemistry	8-25 µg/mL	See Below

DATA

<p>Western Blot</p>	<p>Detection of Human BNIP3 by Western Blot. Western blot shows lysates of MCF-7 human breast cancer cell line untreated (-) or treated (+) with CoCl₂. PVDF membrane was probed with 0.5 µg/mL of Mouse Anti-Human BNIP3 Polyclonal Antibody (Catalog # MAB41471) followed by HRP-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # HAF018). Specific bands were detected for BNIP3 at approximately 25-30 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.</p>	<p>Immunocytochemistry</p> <p>BNIP3 in HeLa Human Cell Line. BNIP3 was detected in immersion fixed HeLa human cervical epithelial carcinoma cell line using Mouse Anti-Human BNIP3 Polyclonal Antibody (Catalog # MAB41471) at 8 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Mouse IgG Secondary Antibody (red; Catalog # NL007) and counterstained with DAPI (blue). Specific staining was localized to cytoplasm. View our protocol for Fluorescent ICC Staining of Cells on Coverslips.</p>
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PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.5 mg/mL in sterile PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
Stability & Storage	Use a manual defrost freezer and avoid repeated freeze-thaw cycles. <ul style="list-style-type: none"> ● 12 months from date of receipt, -20 to -70 °C as supplied. ● 1 month, 2 to 8 °C under sterile conditions after reconstitution. ● 6 months, -20 to -70 °C under sterile conditions after reconstitution.

BACKGROUND

Bcl-2/adenovirus E1B 19 kDa protein-interacting protein 3 (BNIP3), also known as 19 kDa interacting protein 3 (NIP3), is a proapoptotic member of Bcl-2 protein family. BNIP3 is a 194 amino acid, 21.5 kDa (predicted) protein that contains a single Bcl-2 homology 3 (BH3) domain and a C-terminal transmembrane domain required for mitochondrial localization, homodimerization, and regulation of its proapoptotic function. BNIP3 was identified as one of several proteins that interact with discrete domains of Bcl-2 and the E1B 19 kDa protein. Under conditions of prolonged oxygen deprivation, the hypoxia-induced protein HIF1-α activates expression of BNIP3, which in turn, promotes apoptosis under these conditions. The mechanism of BNIP3-mediated apoptosis is independent of caspase activation and cytochrome c release and is characterized by early plasma membrane and mitochondrial damage, prior to the appearance of chromatin condensation or DNA fragmentation. Human BNIP3 shares 90% amino acid identity with mouse and rat BNIP3. Human BNIP3 shares 56% amino acid sequence identity with human BNIP3L.