

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human NFκB2 in direct ELISAs and Western blots. In Western blots, no cross-reactivity with endogenous mouse NFκB2 in NIH 3T3 cells is observed.
Source	Monoclonal Mouse IgG ₁ Clone # 291319
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	<i>E. coli</i> -derived recombinant human NFκB2 Met1-Asn447 Accession # Q00653
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
Chromatin Immunoprecipitation (ChIP)	5 μg/5 x 10 ⁶ cells	See Below
Immunocytochemistry	3-25 μg/mL	See Below

DATA

Western Blot

Detection of Human NFκB2 by Western Blot. Western blot shows lysates of Daudi human Burkitt's lymphoma cell line untreated (-) or treated (+) with 100 ng/mL Recombinant Human CD40 Ligand/TNFSF5 aa 108-261 (Catalog # 6245-CL) for 4 hours. Gels were loaded with 20 μg of cytoplasmic (Cyto) and 10 μg of nuclear extracts (Nuc). For additional reference, lysates of Raji human Burkitt's lymphoma cell line were included. PVDF membrane was probed with 0.5 μg/mL Mouse Anti-Human NFκB2 Monoclonal Antibody (Catalog # MAB28881) followed by HRP-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # HAF007). Specific bands for NFκB2 were detected at approximately 52 kDa and 100 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Chromatin Immunoprecipitation (ChIP)

Detection of NFκB2-regulated Genes by Chromatin Immunoprecipitation. Jurkat human acute T cell leukemia cell line treated with 50 ng/mL PMA and 200 ng/mL calcium ionomycin overnight was fixed using formaldehyde, resuspended in lysis buffer, and sonicated to shear chromatin. NFκB2/DNA complexes were immunoprecipitated using 5 μg Mouse Anti-Human NFκB2 Monoclonal Antibody (Catalog # MAB28881) or control antibody (Catalog # MAB002) for 15 minutes in an ultrasonic bath, followed by Biotinylated Anti-Mouse IgG Secondary Antibody (Catalog # BAF007). Immunocomplexes were captured using 50 μL of MagCollect Streptavidin Ferrofluid (Catalog # MAG999) and DNA was purified using chelating resin solution. The *c-myc* promoter was detected by standard PCR.

Immunocytochemistry

NFκB2 in HeLa Human Cell Line. NFκB2 was detected in immersion fixed HeLa human cervical epithelial carcinoma cell line using Mouse Anti-Human NFκB2 Monoclonal Antibody (Catalog # MAB28881) at 3 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 and nuclei. View our protocol for [Fluorescent ICC Staining of Cells on Coverslips](#).

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

Nuclear Factor kappa B2 (NFκB2 or NFκB p52) is a member of the NFκB/Rel family of transcription factors. NFκB2 dimerizes with other members of the NFκB/Rel family to regulate expression of genes that participate in immune, apoptotic, and oncogenic processes.