Human/Mouse c-Rel Antibody

Antigen Affinity-purified Polyclonal Goat IgG
Catalog Number: AF2699

DESCRIPTION

Species Reactivity	Human/Mouse
Specificity	Detects human and mouse c-Rel in Western blots.
Source	Polyclonal Goat IgG
Purification	Antigen Affinity-purified
Immunogen	*E. coli*-derived recombinant mouse c-Rel Met1-Ile588 Accession # NP_033070
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.

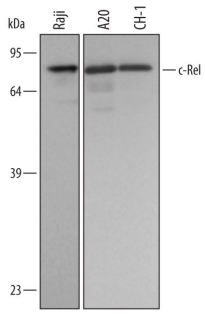
APPLICATIONS

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/10^6 cells	See Below
Immunocytochemistry	5-15 µg/mL	See Below
Simple Western	5 µg/mL	See Below

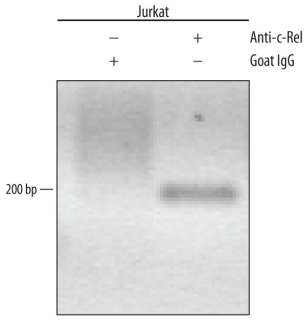
DATA

Western Blot



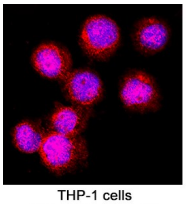
Detection of Human/Mouse c-Rel by Western Blot. Western blot shows lysates of Raji human Burkitt's lymphoma cell line, A20 mouse B cell lymphoma cell line, and CH-1 mouse B cell lymphoma cell line. PVDF membrane was probed with 0.5 µg/mL of Goat Anti-Human/Mouse c-Rel Antigen Affinity-purified Polyclonal Antibody (Catalog # AF2699) followed by HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF109). A specific band was detected for c-Rel at approximately 69 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Chromatin Immunoprecipitation (ChIP)

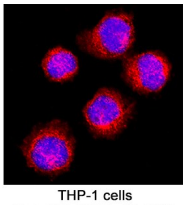


Detection of c-Rel-regulated Genes by Chromatin Immunoprecipitation. Jurkat human acute T cell leukemia cell line treated with 50 ng/mL and 200 ng/mL PMA and calcium ionomycin for overnight was fixed using formaldehyde, resuspended in lysis buffer, and sonicated to shear chromatin. c-Rel/DNA complexes were immunoprecipitated using 5 µg Goat Anti-Human/Mouse c-Rel Antigen Affinity-purified Polyclonal Antibody (Catalog # AF2699) or control antibody (Catalog # AB-108-C) for 15 minutes in an ultrasonic bath, followed by Biotinylated Anti-Goat IgG Secondary Antibody (Catalog # BAF109). Immunocomplexes were captured using 50 µL of MagCellect Streptavidin Ferrofluid (Catalog # MAG999) and DNA was purified using chelating resin solution. The *p21* promoter was detected by standard PCR.

Immunocytochemistry



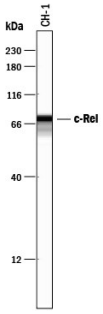
THP-1 cells
Stimulated with LPS



THP-1 cells
Non-stimulated with LPS

c-Rel in THP-1 Human Cell Line. c-Rel was detected in immersion fixed THP-1 human acute monocytic leukemia cell line treated with LPS using Goat Anti-Human/Mouse c-Rel Antigen Affinity-purified Polyclonal Antibody (Catalog # AF2699) at 5 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Goat IgG Secondary Antibody (red; Catalog # NL001) and counterstained with DAPI (blue). Specific staining was localized to cell nuclei upon stimulation with LPS. View our protocol for Fluorescent ICC Staining of Non-adherent Cells.

Simple Western



Detection of Mouse c-Rel by Simple Western™. Simple Western lane view shows lysates of CH-1 mouse B cell lymphoma cell line, loaded at 0.2 mg/mL. A specific band was detected for c-Rel at approximately 71 kDa (as indicated) using 5 µg/mL of Goat Anti-Human/Mouse c-Rel Antigen Affinity-purified Polyclonal Antibody (Catalog # AF2699) followed by 1:50 dilution of HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF109). This experiment was conducted under reducing conditions and using the 12-230 kDa separation system.

bio-techne®
Global bio-techne.com info@bio-techne.com techsupport@bio-techne.com TEL +1 612 379 2956
USA TEL 800 343 7475 Canada TEL 855 668 8722 China TEL +86 (21) 52380373
Europe | Middle East | Africa TEL +44 (0)1235 529449

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Antigen Affinity-purified Polyclonal Goat IgG

Catalog Number: AF2699

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.2 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.** • 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

c-Rel/Rel, the cellular counterpart of the v-Rel oncogene of the avian reticuloendotheliosis retrovirus, is a 69 kDa class II member of the Rel/NK-κB family of transcription factors. All Rel family members contain a RHD (Rel homology domain) that is involved in dimerization, DNA and IκB binding, and nuclear localization. Class II members contain an additional C-terminal transcriptional activation segment. Rel both homodimerizes and heterodimerizes with multiple family members. Following dimerization, Rel complexes initiate transcription by acting on decameric DNA motifs termed κB binding sites. The important role of c-Rel in B-cell development, growth, and survival is well documented. c-Rel is also involved in responses to auto-antigens, allo-antigens, allergens and pathogens and may contribute to the development of certain human lymphoid cell cancers.

Global bio-techne.com info@bio-techne.com techsupport@bio-techne.com TEL +1 612 379 2956
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Europe | Middle East | Africa TEL +44 (0)1235 529449