Human c-Myc Antibody

Antigen Affinity-purified Polyclonal Goat IgG
Catalog Number: AF3696

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human c-Myc in direct ELISAs and Western blots. In direct ELISAs, less than 1% cross-reactivity with recombinant human (rh) L-Myc and rhN-Myc is observed.
Source	Polyclonal Goat IgG
Purification	Antigen Affinity-purified
Immunogen	*E. coli*-derived recombinant human c-Myc Arg66-Asp201 Accession # P01106
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 as a 0.2 μm filtered solution in PBS.

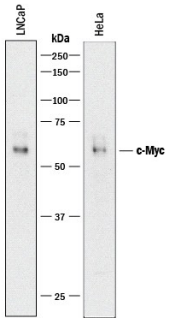
APPLICATIONS

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	5-15 µg/mL	See Below
Immunohistochemistry	5-15 µg/mL	Immersion fixed paraffin-embedded sections of human liver cancer tissue
Simple Western	20 µg/mL	See Below

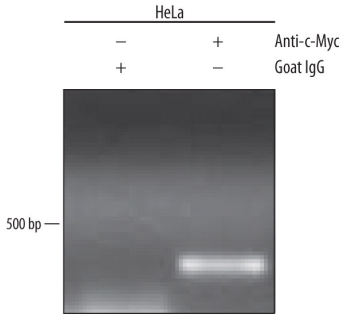
DATA

Western Blot



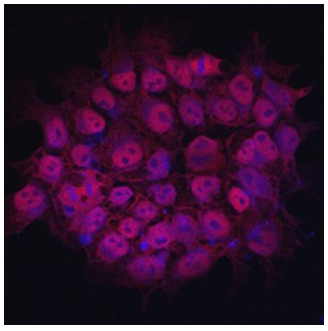
Detection of Human c-Myc by Western Blot. Western blot shows lysates of LNCaP human prostate cancer cell line and HeLa human cervical epithelial carcinoma cell line. PVDF membrane was probed with 0.5 µg/mL of Goat Anti-Human c-Myc Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3696) followed by HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF017). A specific band was detected for c-Myc at approximately 56 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Chromatin Immunoprecipitation (ChIP)



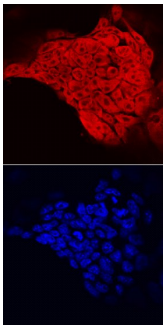
Detection of c-Myc-regulated Genes by Chromatin Immunoprecipitation. HeLa human cervical epithelial carcinoma cell line was fixed using formaldehyde, resuspended in lysis buffer, and sonicated to shear chromatin. c-Myc/DNA complexes were immunoprecipitated using 5 µg Goat Anti-Human c-Myc Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3696) 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



c-Myc in BG01V Human Stem Cells. c-Myc was detected in immersion fixed BG01V human embryonic stem cells using 10 µg/mL Goat Anti-Human c-Myc Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3696) for 3 hours at room temperature. Cells were stained with the NorthernLights™ 557-conjugated Anti-Goat IgG Secondary Antibody (red; Catalog # NL001) and counterstained with DAPI (blue). View our protocol for Fluorescent ICC Staining of Cells on Coverslips.

Immunocytochemistry



c-Myc in D3 Mouse Stem Cells. c-Myc was detected in immersion fixed D3 mouse embryonic stem cell line using Goat Anti-Human c-Myc Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3696) at 10 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Goat IgG Secondary Antibody (red, upper panel; Catalog # NL001) and counterstained with DAPI (blue, lower panel). Specific staining was localized to nuclei and cytoplasm. View our protocol for Fluorescent ICC Staining of Cells on Coverslips.

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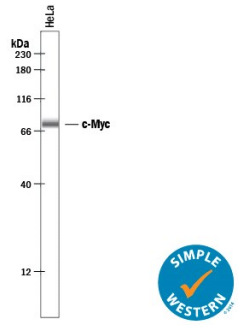
Simple Western



Detection of Human c-Myc by Simple Western™. Simple Western lane view shows lysates of HeLa human cervical epithelial carcinoma cell line, loaded at 0.2 mg/mL. A specific band was detected for c-Myc at approximately 78 kDa (as indicated) using 20 µg/mL of Goat Anti-Human c-Myc Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3696) 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.

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

Human c-Myc is a 439 amino acid transcription factor with a bHLH/LZ (basic Helix-Loop-Helix, Leucine Zipper) domain. c-Myc DNA-binding and transcription function is achieved upon heterodimerization with its partner Max. c-Myc is often over-expressed and mutated in hematopoietic tumors. Mutations frequently result in truncations that remove the transactivation region or in the bHLH/LZ domain required for association with Max and DNA. Over the region used as immunogen, human c-Myc is 92% identical to the rat and mouse c-Myc proteins.

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