

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human KLF4 in direct ELISAs and Western blots.
Source	Polyclonal Goat IgG
Purification	Antigen Affinity-purified
Immunogen	<i>E. coli</i> -derived recombinant human KLF4 Ala2-Phe470 Accession # AAH29923
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.

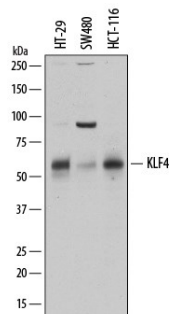
APPLICATIONS

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	5-15 µg/mL	See Below
Immunohistochemistry	15 µg/mL	See Below
Simple Western	25 µg/mL	See Below

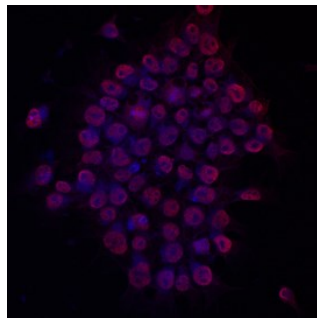
DATA

Western Blot



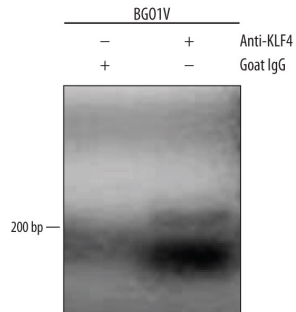
Detection of Human KLF4 by Western Blot. Western blot shows lysates of HT-29 human colon adenocarcinoma cell line, SW480 human colorectal adenocarcinoma cell line, and HCT-116 human colorectal carcinoma cell line. PVDF membrane was probed with 0.5 µg/mL of Goat Anti-Human KLF4 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3640) followed by HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF109). A specific band was detected for KLF4 at approximately 60 kDa (as indicated). This experiment was conducted under reducing conditions and using [Immunoblot Buffer Group 5](#).

Immunocytochemistry



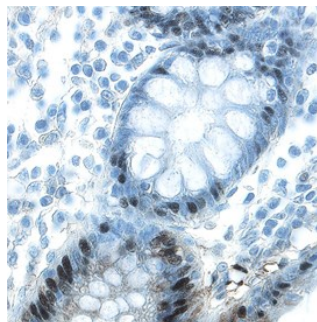
KLF4 in BG01V Human Stem Cells. KLF4 was detected in immersion fixed BG01V human embryonic stem cells using 10 µg/mL Goat Anti-Human KLF4 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3640) for 3 hours at room temperature. Cells were stained with the NorthernLights™ 557-conjugated Anti-Goat IgG Secondary Antibody (red; Catalog # NL001) and counterstained with DAPI (blue). View our protocol for [Fluorescent ICC Staining of Cells on Coverslips](#).

Chromatin Immunoprecipitation (ChIP)



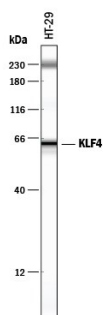
Detection of KLF4-regulated Genes by Chromatin Immunoprecipitation. BG01V human embryonic stem cells were fixed using formaldehyde, resuspended in lysis buffer, and sonicated to shear chromatin. KLF4/DNA complexes were immunoprecipitated using 5 µg Goat Anti-Human KLF4 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3640) 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 MagCelect Streptavidin Ferrofluid (Catalog # MAG999) and DNA was purified using chelating resin solution. The B2R promoter was detected by standard PCR.

Immunohistochemistry



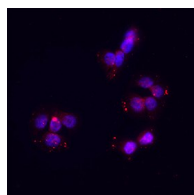
KLF4 in Human Colon. KLF4 was detected in immersion fixed paraffin-embedded sections of human colon using 15 µg/mL Goat Anti-Human KLF4 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3640) overnight at 4 °C. Tissue was stained with the Anti-Goat HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # CTS008) and counterstained with hematoxylin (blue). Specific labeling was localized to the nuclei of epithelial cells. View our protocol for Chromogenic IHC Staining of Paraffin-embedded Tissue Sections.

Simple Western

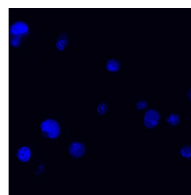


Detection of Human KLF4 by Simple Western™. Simple Western lane view shows lysates of HT-29 human colon adenocarcinoma cell line, loaded at 0.2 mg/mL. A specific band was detected for KLF4 at approximately 63 kDa (as indicated) using 25 µg/mL of Goat Anti-Human KLF4 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3640) 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. Non-specific interaction with the 230 kDa Simple Western standard may be seen with this antibody.

Immunocytochemistry



HT-29 (Positive) cells



HDLM-2 (Negative) cells

Detection of KLF4 in HT-29 Human Colon Adenocarcinoma Cell Line (positive) and HDLM-2 Human Hodgkin's Lymphoma Cell Line (negative) Cells. KLF4 was detected in immersion fixed HT-29 Human Colon Adenocarcinoma Cell Line (positive) and HDLM-2 Human Hodgkin's Lymphoma Cell Line (negative) Cells using Goat Anti-Human KLF4 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3640) 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 cytoplasm. View our protocol for Fluorescent ICC Staining of Cells on Coverslips.

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. <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

Human KLF4, also known as epithelial zinc finger protein (EZP), is a 53 kDa (470 aa) member of the kruppel C2H2-type zinc finger protein family. It contains three C2H2-type zinc fingers at the carboxyl terminus that preferentially bind to cis-DNA elements that are GC rich. KLF4 regulates the expression of target genes that are involved in different cellular functions. KLF4 is highly expressed in the epithelial cells of the skin and the gastrointestinal tract. Human and mouse KLF4 share 90% amino acid sequence identity.