Human GLI-1 Antibody

Antigen Affinity-purified Polyclonal Goat IgG
Catalog Number: AF3324

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human GLI-1 in direct ELISAs and Western blots. In Western blots, approximately 5% cross-reactivity with recombinant human GLI-3 is observed.
Source	Polyclonal Goat IgG
Purification	Antigen Affinity-purified
Immunogen	*E. coli*-derived recombinant human GLI-1 Met1-Glu234 Accession # P08151
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.

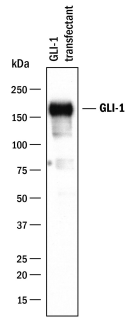
APPLICATIONS

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	1 µg/mL	See Below
Chromatin Immunoprecipitation (ChIP)	5 µg/10^6 cells	See Below
Immunohistochemistry	5-15 µg/mL	See Below

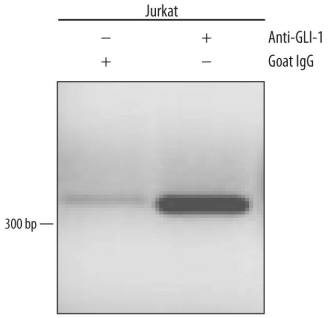
DATA

Western Blot



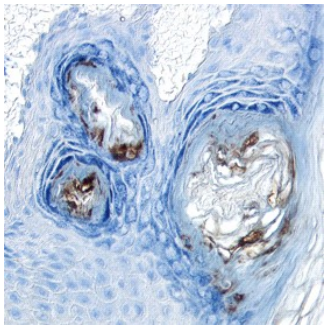
Detection of Human GLI-1 by Western Blot. Western blot shows lysates of HEK293 human embryonic kidney cell line transfected with human GLI-1. PVDF membrane was probed with 1 µg/mL of Goat Anti-Human GLI-1 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3324) followed by HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF017). A specific band was detected for GLI-1 at approximately 165 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Chromatin Immunoprecipitation (ChIP)



Detection of GLI-1-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 for 30 mintes were fixed using formaldehyde, resuspended in lysis buffer, and sonicated to shear chromatin. GLI-1/DNA complexes were immunoprecipitated using 5 µg Goat Anti-Human GLI-1 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3324) 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 *Bcl-2* promoter was detected by standard PCR.

Immunohistochemistry



GLI-1 in Human Skin. GLI-1 was detected in immersion fixed paraffin-embedded sections of human skin using Human GLI-1 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3324) at 5 µg/mL overnight at 4 °C. Tissue was stained using the Anti-Goat HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # CTS008) and counterstained with hematoxylin (blue). View our protocol for Chromogenic IHC Staining of Paraffin-embedded Tissue Sections.

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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

GLI-1 is a transcription regulator with 5 conserved tandem C2-H2 zinc finger domains, joined by conserved histidine/cysteine linkers. It is a known oncogene that was originally isolated in a human glioblastoma. GLI-1 acts downstream of Hedgehog signaling and is involved in cell proliferation and pattern formation during embryonic development. Within the region used as the immunogen, human GLI-1 shares 81% amino acid sequence homology with mouse GLI-1.

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