Datasheet for 600-401-695

Gli2 Antibody

Overview

Description:	Anti-Gli2 (RABBIT) Antibody - 600-401-695
Item No.:	600-401-695
Size:	100 µg
Applications:	ELISA, IHC, WB, IF
Reactivity:	Mouse
Host Species:	Rabbit

Product Details

Background:	Gli-2 (also known as Zinc Finger Protein Gli-2, GLI-Kruppel family member GLI-2 or Tax helper protein) belongs to the C2H2-type zinc finger protein subclass of the Gli family. Members of this subclass are characterized as transcription factors that bind DNA through zinc finger motifs. These motifs contain conserved H-C links. Gli family zinc finger proteins are mediators of Sonic hedgehog (Shh) signaling, and they are implicated as potent oncogenes in the embryonal carcinoma cell. The protein encoded by this gene localizes to the cytoplasm and activates patched Drosophila homolog (PTCH) gene expression. Gli-2 is also thought to play a role during embryogenesis. The encoded protein is associated with several phenotypes: Greig cephalopolysyndactyly syndrome, Pallister-Hall syndrome, pre-axial polydactyly type IV, and postaxial polydactyly types A1 and B. Anti-Gli 2 Antibody is useful for researchers interested in transcription factor activities, DNA binding, and chromatin binding research.
Synonyms:	rabbit anti-Gli-2 antibody, Gli 2, Gli2, zinc finger protein GLI2, Tax helper protein antibody, Thp antibody
Host Species:	Rabbit
Clonality:	Polyclonal
Format:	IgG

Target Details

Gene Name:	Gli2
Reactivity:	Mouse

Immunogen Type:	Conjugated Peptide
Immunogen:	This affinity purified antibody was prepared from whole rabbit serum produced by repeated immunizations with a synthetic peptide corresponding to amino acids from an internal region of Mouse Gli-2.
Purity/Specificity:	This affinity purified antibody is directed against mouse Gli-2 protein. The product was affinity purified from monospecific antiserum by immunoaffinity chromatography. A BLAST analysis was used to suggest cross-reactivity with Gli-2 from mouse and rat sources based on 100% sequence homology with the immunogen. Reactivity with Gli-2 from other sources is not known.
Relevant Links:	• UniProtKB - Q8K0K3 • NCBI - 124487481 • GeneID - 14633

Application Details

Tested Applications:	ELISA, IHC, WB
Suggested Applications:	IF (Based on references)
Application Note:	This antibody has been tested for use in ELISA, immunohistochemistry and western blot. Specific conditions for reactivity should be optimized by the end user. See figure legend for expectations by WB and IHC. Multiple splice variants have been reported for this protein.
Assay Dilutions:	All assays should be optimized by the user. Recommended dilutions (if any) may be listed below.
ELISA:	1:15,000 - 1:60,000
IHC:	2 µg/ml to 20 µg/ml
WB:	1:500 - 1:2,000

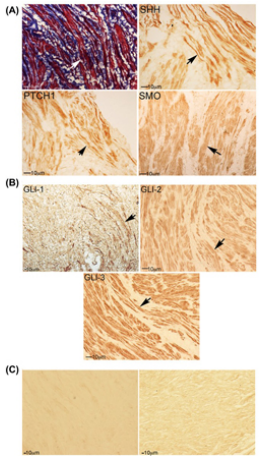
Formulation

Physical State:	Liquid (sterile filtered)
Concentration:	1.02 mg/mL by UV absorbance at 280 nm
Buffer:	0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2
Preservative:	0.01% (w/v) Sodium Azide
Stabilizer:	None

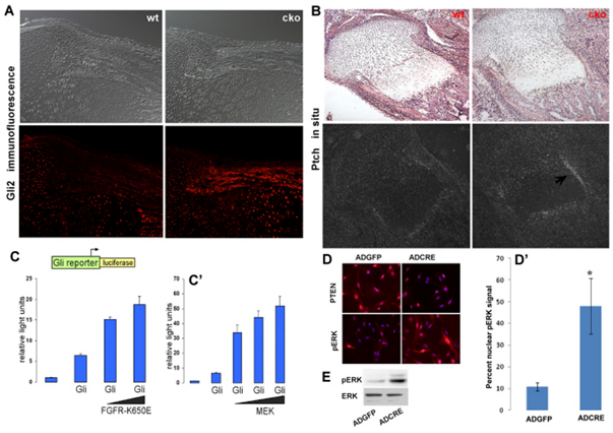
Shipping & Handling

Shipping Condition:	Dry Ice
Storage Condition:	Store vial at -20° C prior to opening. Aliquot contents and freeze at -20° C or below for extended storage. Avoid cycles of freezing and thawing. Centrifuge product if not completely clear after standing at room temperature. This product is stable for several weeks at 4° C as an undiluted liquid. Dilute only prior to immediate use.
Expiration:	Expiration date is one (1) year from date of receipt.

Images

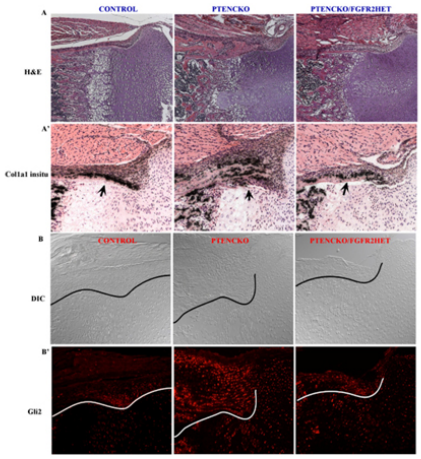


Immunohistochemistry
Trichrome stain and IHC analysis for SHH pathway in human RS muscle. A, Trichrome stain of human RS indicates abundant muscle and collagen. Arrows indicate muscle. IHC analysis of human RS shows SHH, PTCH1, and SMO protein localization in RS muscle. B, GLI-1, GLI-2 (p/n 600-401-695), and GLI-3 (p/n 600-401-694) are abundant in RS muscle. C, No primary controls (left: mouse anti-rabbit, and right: donkey anti-goat) are presented. Arrows indicate protein. 100–200× magnification. Fig 1. PMID: 30187971

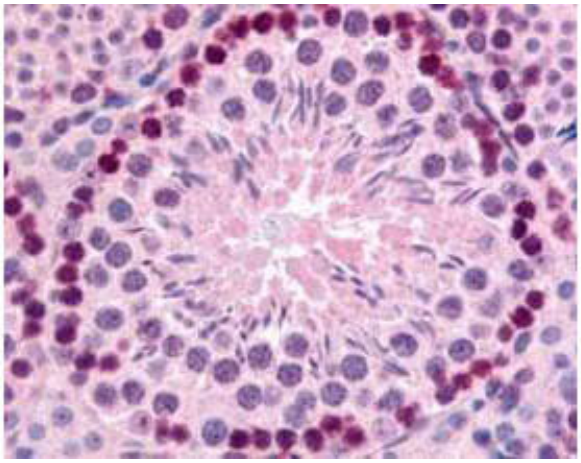


Immunofluorescence Microscopy

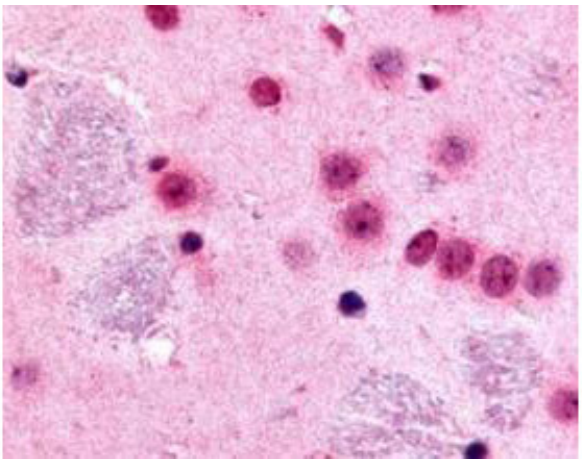
Effect of Pten deletion on hedgehog signaling. (A) Immunofluorescence of GLI2 showed an increase in the expression of osteoblasts that line the perichondrium in the Pten conditional knockout (cko) compared with wild type (wt). (B) In situ hybridization for patched (Ptch1), a transcriptional target for GLI2, showed an increase (black arrow) in Ptch1 expression in the cko compared with the wt control. (C,C') Using a Gli2 luciferase reporter plasmid transfected into C3H10T1/2 cells we observed that there is an activation of Gli2 transcription when an active form of FGFR (FGFR-K650E) is co-transfected with GLI2. We also observed an increase in Gli2 luciferase activity when we used an activated MEK kinase plasmid (repeated at least three times). Black wedges indicate increasing concentrations of FGFR-K650E or MEK co-transfected with Gli2. Luciferase activity was normalized to β-galactosidase activity. (D) Using Pten flox/flox calvarial osteoblasts, we used AdenoGFP (ADGFP) as control and AdenoCRE (ADCRE) to delete PTEN as can be observed in the indirect immunofluorescence images in the top panels. When PTEN is deleted we observe an increase in pERK nuclear localization. (D') The number of cells with a nuclear pERK signal was determined revealing a significant increase in pERK (n=3, *P<0.005). Images are overlays of DAPI (blue) and rhodamine (red) staining. (E) Western blot data for pERK and total ERK from protein lysates of Pten flox/flox calvarial osteoblasts treated with AdenoCRE and AdenoGFP. All error bars indicate s.d. Fig 7. PMID: 21385768



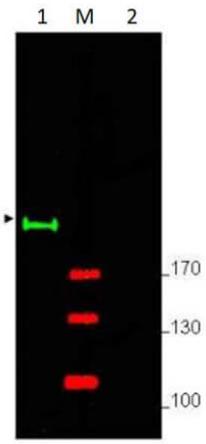
Immunohistochemistry
Rescue of the Pten conditional knockout (cko) mouse phenotype by deletion of one allele of Fgfr2. (A,A') Hematoxylin and Eosin (H&E) staining of the phenotype observed in the perichondrium, along with Col1a1 in situ hybridization showing the increased perichondrium (black arrows) phenotype in the absence of Pten (PTENCKO). This phenotype is partially rescued with the deletion of Pten in the background of global loss of one allele of Fgfr2 (PTENCKO/FGFR2HET). (B,B') Immunohistochemistry for GLI2 protein levels in the perichondrium of tibial sections showed a similar increase in GLI2 protein levels in the absence of Pten but the GLI2 levels are comparable to the wild-type levels in the Fgfr2 het Pten cko (PTENCKO/FGFR2HET) perichondrium. The conditional knockouts were generated using the Dermo1cre mouse strain. DIC, differential interference contrast. Fig 8.
PMID: 21385768



Immunohistochemistry
Rockland's Affinity Purified anti-mouse Gli-2 antibody was used at 10 µg/ml to evaluate staining on several mouse tissues. Moderate to strong staining was seen on many tissues, with low background staining. This image shows Gli-2 staining of mouse testis. Tissue was formalin-fixed and paraffin embedded.



Immunohistochemistry
Rockland's Affinity Purified anti-mouse Gli-2 antibody was used at 10 µg/ml to evaluate staining on several mouse tissues. Moderate to strong staining was seen on many tissues with low background staining. This image shows Gli-2 staining of mouse brain. Tissue was formalin-fixed and paraffin embedded.



Western Blot
Western blot using Rockland's Affinity Purified anti-Gli-2 antibody shows detection of a predominant band at ~190 kDa corresponding to Gli-2 (arrowhead) in mouse brain whole cell lysate (p/n W10-000-T004) (lane 1). Pre-incubation of antibody with immunizing peptide completely blocks staining of this band (lane 2). Load 25µg of lysate was resolved on a 4-8% Tris-glycine gel by SDS-PAGE and transferred onto nitrocellulose. After blocking with 5% goat serum and 0.5% BLOTTO in PBS, the membrane was probed with the primary antibody diluted to 1:750. Incubation was at room temperature for 2 h followed by washes and reaction with a 1:10,000 dilution of IRDye® 800 conjugated Gt-a-Rabbit IgG (H&L) MX10 (p/n 611-132-122) for 45 min at room temperature. Molecular weight markers are shown (M) using the 700 nm channel (red). IRDye® 800 fluorescence image was captured using the Odyssey® Infrared Imaging System developed by LI-COR. IRDye is a trademark of LI-COR, Inc. Other detection systems will yield similar results.

References

- Hehemann M et al. Sonic hedgehog regulation of human rhabdosphincter muscle:Potential implications for treatment of stress urinary incontinence. *Neurourol Urodyn.* (2018)
- Guntur AR et al. Conditional ablation of Pten in osteoprogenitors stimulates FGF signaling. *Development.* (2011)

Disclaimer

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