

PRODUCT INFORMATION

Product name : GLI1 antibody

Product type : Primary antibodies

Description : Rabbit polyclonal to GLI1

Immunogen : 3 synthetic peptides (human) conjugated to KLH

Reacts with : Human, Mouse

Tested applications : ELISA, WB and IF

GENE INFORMATION

Gene Symbol : GLI1

Gene Name : GLI family zinc finger 1

Ensembl ID : ENSG00000111087

Entrez GeneID : 2735

Omim ID : 165220

Swiss-Prot : P08151

Molecular weight of GLI1 : 117.9kDa

Function : Acts as a transcriptional activator. May regulate the transcription of specific genes during normal development. May play a role in craniofacial development and digital development, as well as development of the central nervous system and gastrointestinal tract. Mediates SHH signaling and thus cell proliferation and differentiation

Expected subcellular localization : Cytoplasm. Nucleus. Note: Tethered in the cytoplasm by binding to SUFU. Activation and translocation to the nucleus is promoted by interaction with STK36. Phosphorylation by ULK3 may promote nuclear localization. Translocation to the nucleus is promoted by interaction with ZIC1

Summary : This gene encodes a member of the Kruppel family of zinc finger proteins. The encoded transcription factor is activated by the sonic hedgehog signal transduction cascade and regulates stem cell proliferation. The activity and nuclear localization of this protein is negatively regulated by p53 in an inhibitory loop. Multiple transcript variants encoding different isoforms have been found for this gene. [provided by RefSeq, May 2009]

APPLICATION NOTE

Recommended dilution :

- **ELISA:** Antibody specificity was verified by direct ELISA against the 3 immunogen peptides. A minimum titer of 1/6000 is determined. Appropriate specificity controls were run.
- **WB (recombinant protein):** 1/1000.
- **WB (cell lysate):** 1/250.
- **IF:** 1/100.

Optimal dilutions/concentration should be determined by the end user.

Raised in : Rabbit

Clonality : Polyclonal

Isotype : IgG

Purity : Purified Antibody

Storage buffer : 0.5 X PBS, 50% glycerol containing a final concentration of 0.1% BSA and 0.01% Thimerosal.

Form : Liquid

Storage instruction : Store at -20°C or -80°C. Avoid repeated freeze / thaw cycles.

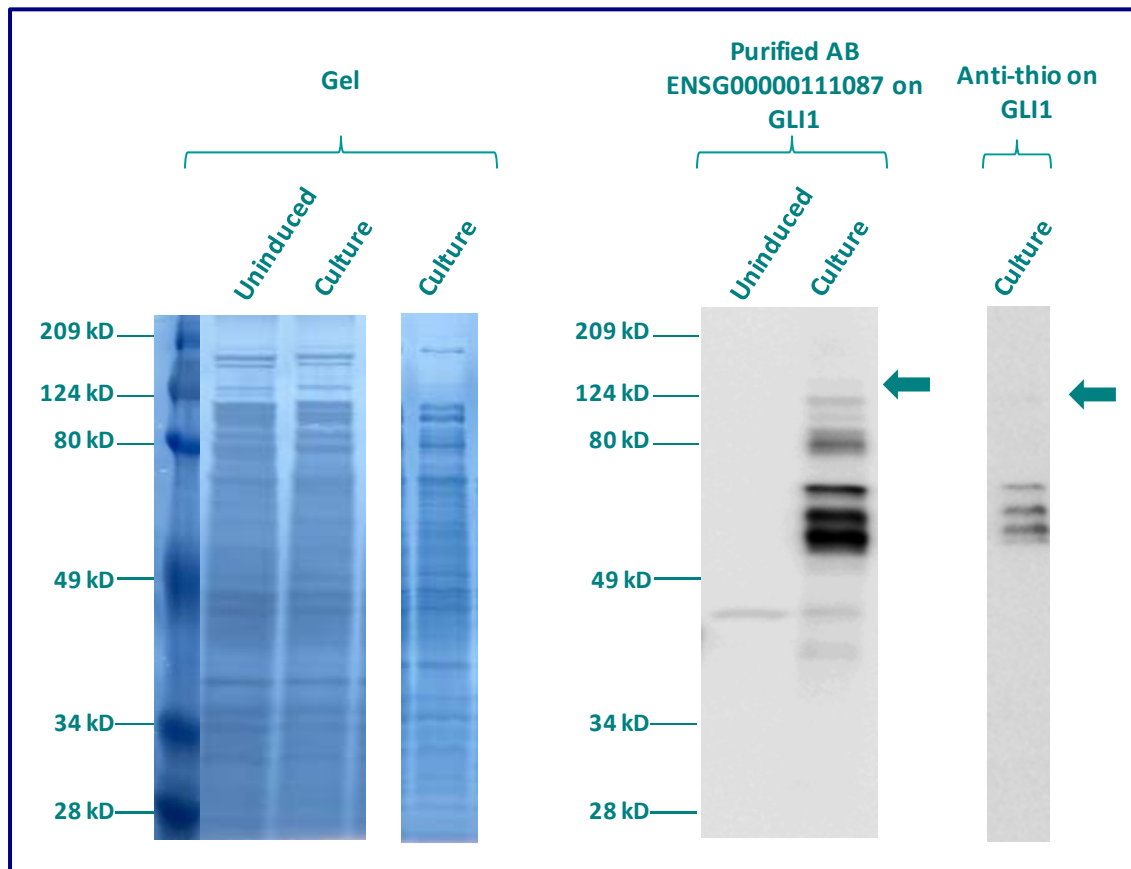
WESTERN BLOT ON RECOMBINANT PROTEIN

The purified antibody ENSG00000111087 has been tested at 1/1000 on uninduced (negative control) and induced culture of E.coli (one shot Top10 competent cells).

An anti-thio has been tested at 1/5000 on induced culture of E.coli (one shot Top10 competent cells) as a positive control.

Plasmid name : pBAD-DEST49.

Molecular weight of GLI1 : 131.9kDa (117.9kDa + another 14kDa for the tag).



Gel concentration: 10%

Blocking: in 5% non-fat milk-PBST solution

1st Antibody: The antibodies are diluted in blocking buffer.

- Dilute the purified antibody ENSG00000111087 at 1: 1000
- Dilute the anti-thio at 1:5000

60 minutes of incubation

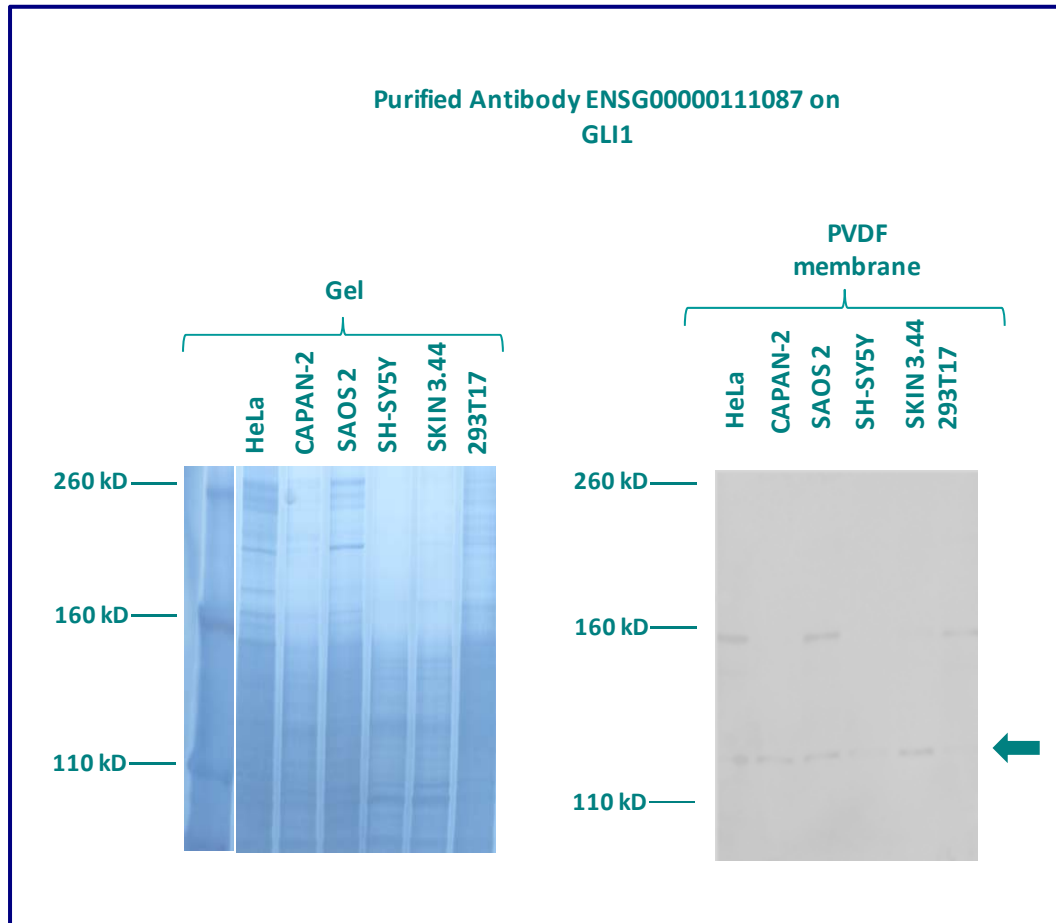
2nd Antibody: The antibody is diluted in blocking buffer.

- Dilute the anti-Rabbit IgG HRP conjugated at 1/10000
- 60 minutes of incubation

WESTERN BLOT ANALYSIS ON CELL LYSATES

The purified antibody ENSG00000111087 has been tested at a concentration of 1/500 on total protein extract of various cell lines (HeLa, CAPAN-2, SAOS 2, SH-SY5Y, SKIN 3.44 & 293T17).

Molecular weight of GLI1 isoforms : 117.9 & 113.7kDa



Gel concentration: 5%

Blocking: in 5% non-fat milk-PBST solution

1st Antibody: The antibodies are diluted in blocking buffer.

- Dilute the purified antibody ENSG00000111087 at 1: 250
- 60 minutes of incubation**

2nd Antibody: The antibody is diluted in blocking buffer.

- Dilute the anti-Rabbit IgG HRP conjugated at 1/10000
- 60 minutes of incubation**

IMMUNOFLUORESCENCE ANALYSIS

Immunofluorescence analysis of Zinc finger protein GLI1 (GLI1) expression in 5 cells lines (HELA, Capan-2, SAOS-2, SH-SY5Y, Skin 3,44). The purified Antibody ENSG00000111087 has been tested at 1/5000.

Red staining : cytoskeleton (microtubules/ α -tubuline)

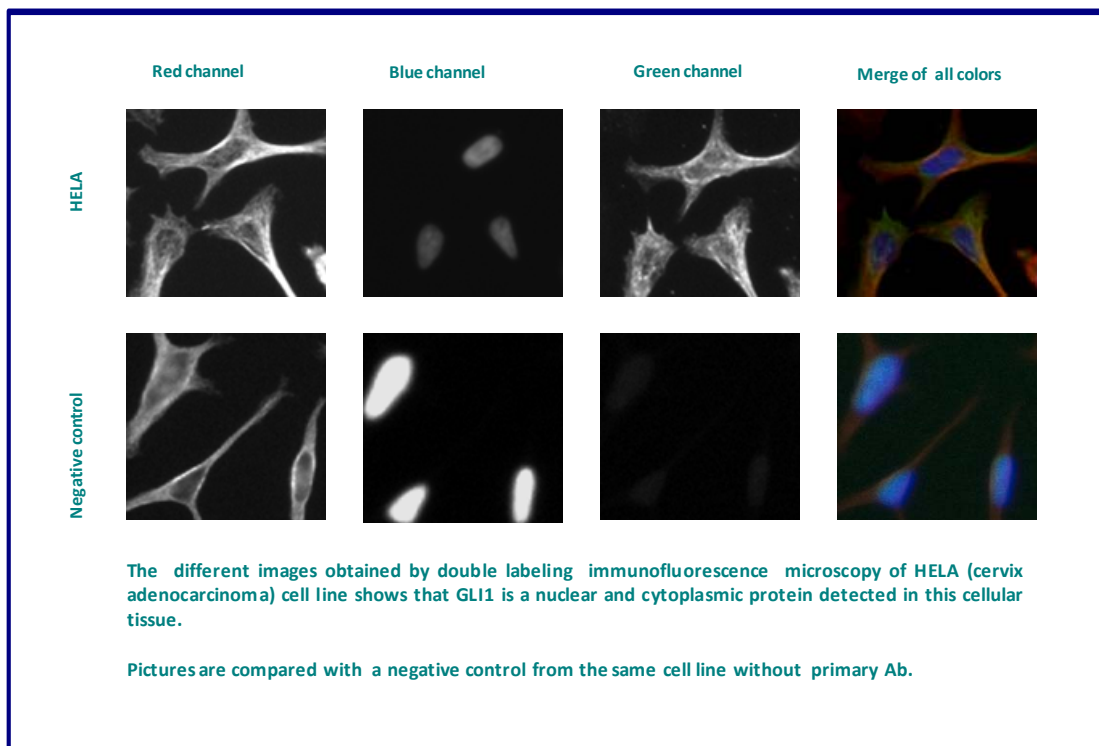
Blue staining : nucleus (Hoechst)

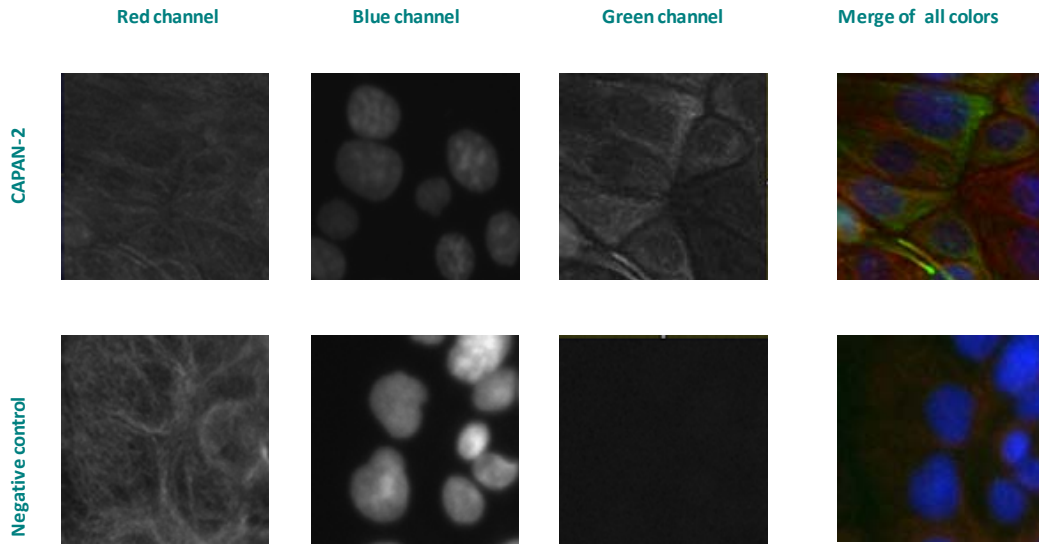
Green staining : anti- GLI1 antibody (purified)

Expected subcellular location : Cytoplasm. Nucleus.

Note: Tethered in the cytoplasm by binding to SUFU. Activation and translocation to the nucleus is promoted by interaction with STK36. Phosphorylation by ULK3 may promote nuclear localization. Translocation to the nucleus is promoted by interaction with ZIC1

Expected tissue specificity : Testis, myometrium and fallopian tube. Also expressed in the brain with highest expression in the cerebellum, optic nerve and olfactory tract.





The different images obtained by double labeling immunofluorescence microscopy of CAPAN- 2 (Pancreas adenocarcinoma) cell line shows that GLI1 is a nuclear and cytoplasmic protein detected in this cellular tissue.

Pictures are compared with a negative control from the same cell line without primary Ab.

Remaining cell lines tested gave a positive result with a nuclear and cytoplasmic distribution.