PRODUCT INFORMATION

Product name：ID2 antibody

Product type： Primary antibodies

Description： Rabbit polyclonal to ID2

Immunogen：3 synthetic peptides (human) conjugated to KLH

Reacts with： Hu, Ms

Tested applications： ELISA, WB and IF

GENE INFORMATION

Gene Symbol：ID2

Gene Name：inhibitor of DNA binding 2, dominant negative helix-loop-helix protein

Ensembl ID：ENSG00000115738

Entrez GeneID：3398

GenBank Accession number：N/A

Omim ID：600386

Swiss-Prot：Q02363

Molecular weight of ID2：14.9 kDa

Function： ID (inhibitor of DNA binding) HLH proteins lack a basic DNA-binding domain but are able to form heterodimers with other HLH proteins, thereby inhibiting DNA binding. ID-2 may be an inhibitor of tissue-specific gene expression.

Expected subcellular localization： Nucleus

Expected tissue specificity： Highly expressed in early fetal tissues, including those of the central nervous system.

Summary： The protein encoded by this gene belongs to the inhibitor of DNA binding (ID) family, members of which are transcriptional regulators that contain a helix-loop-helix (HLH) domain but not a basic domain. Members of the ID family inhibit the functions of basic helix-loop-helix transcription factors in a dominant-negative manner by suppressing their heterodimerization partners through the HLH domains. This protein may play a role in negatively regulating cell differentiation. A pseudogene has been identified for this gene. [provided by RefSeq]

APPLICATION NOTE

Recommended dilution：

- **ELISA: Antibody specificity was verified by direct ELISA against the 3 immunogen peptides. A titer of 1/19000 has been determined. Appropriate specificity controls were run.**
- **WB: 1/5000.**
- **IF: 1/1000.**

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, containing a final concentration of 50% Glycerol, 0.1% BSA and 0.01% Thimerosal.

Form： Liquid

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

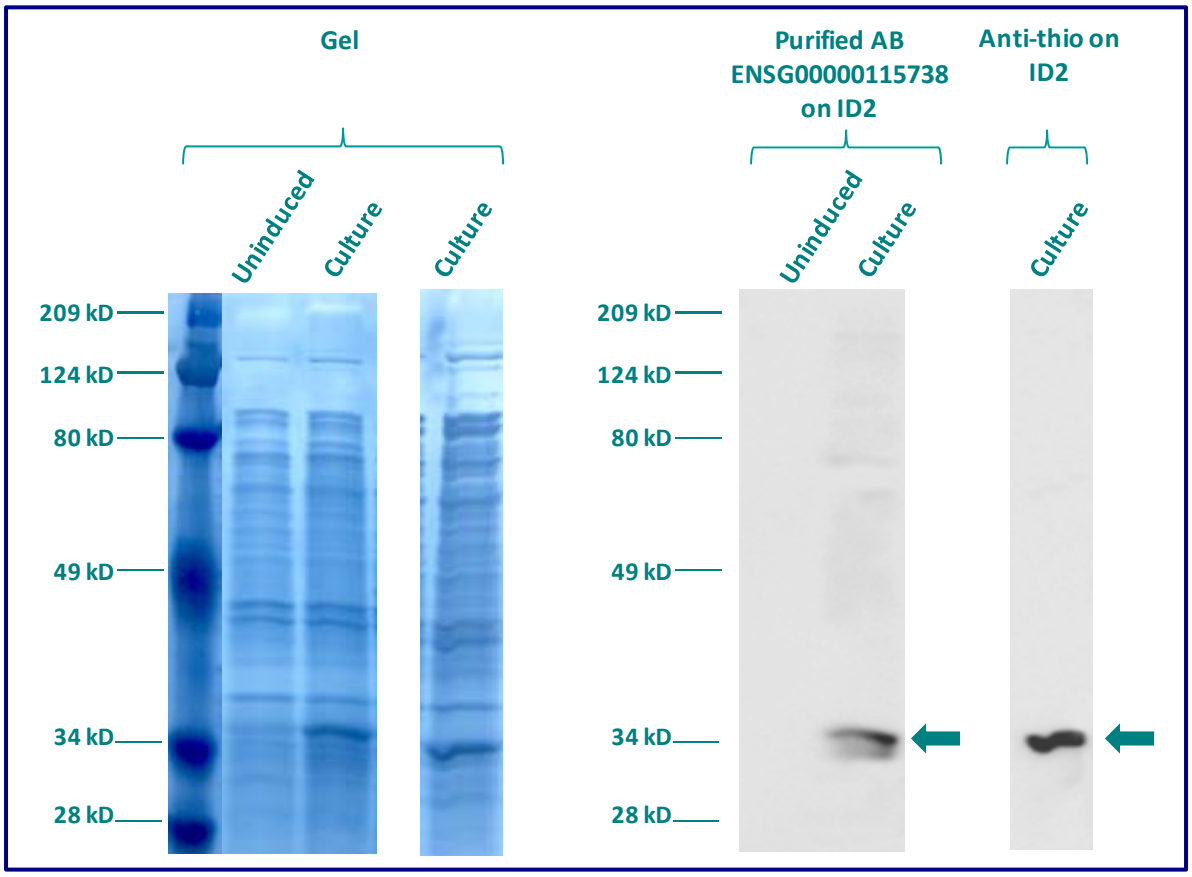
WESTERN BLOT ON RECOMBINANT PROTEIN

The purified antibody ENSG00000115738 has been tested at 1/5000 on uninduced (negative control) and induced culture of E.coli (one shot Top10 competent cells).

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

Plasmid name： pBAD-DEST49.

Molecular weight of ID2 ： 28.9kDa (14.9kDa + 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 ENSG00000115738 at 1:5000**
- **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

IMMUNOFLUORESCENCE ANALYSIS

Immunofluorescence analysis of DNA-binding protein inhibitor ID-2 (ID2) expression in 5 cells lines (HELA, Capan-2, SAOS-2, SH-SY5Y, Skin 3,44). The purified Antibody ENSG00000115738 has been tested at 1/5000.

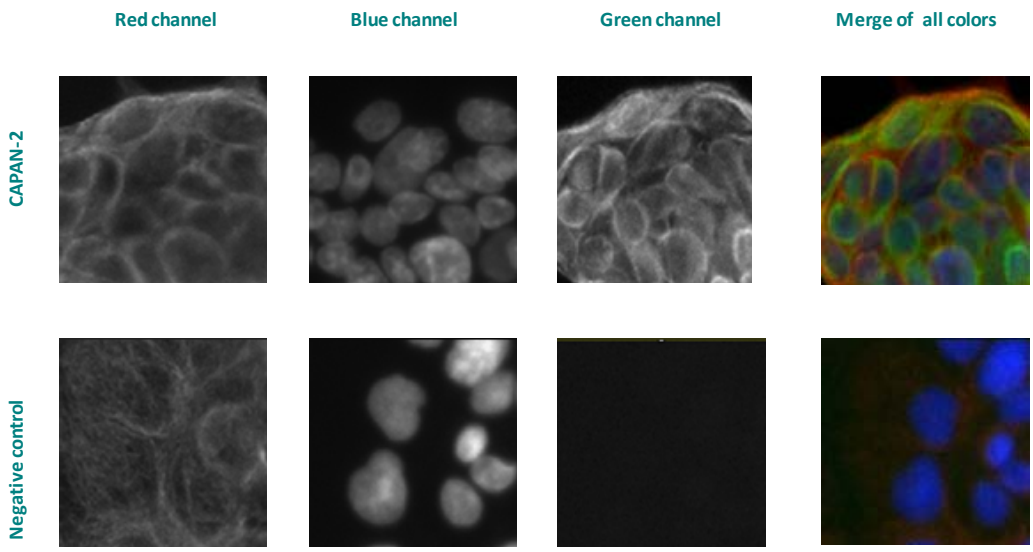
Red staining : cytoskeleton (microtubules/α-tubuline)

Blue staining : nucleus (Hoechst)

Green staining : anti- ID2 antibody (purified)

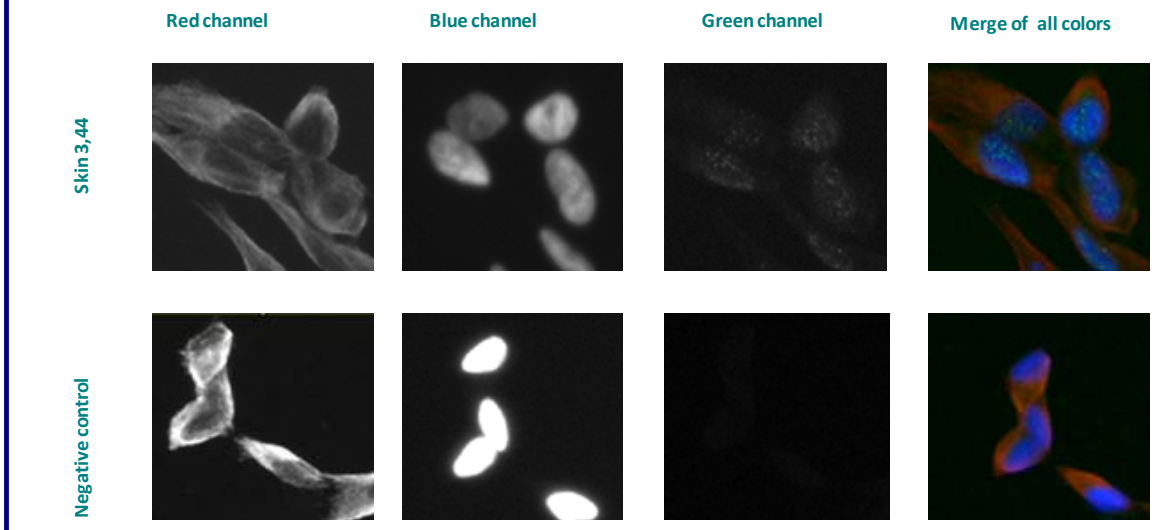
Expected subcellular location : Cytoplasm. Nucleus

Expected tissue specificity : Highly expressed in early fetal tissues, including those of the central nervous system



The different images obtained by double labeling immunofluorescence microscopy of CAPAN- 2 (Pancreas adenocarcinoma) cell line shows that ID2 is a cytoplsmic protein detected in this cellular tissue.

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



The different images obtained by double labeling immunofluorescence microscopy of Skin 3,44 (melanoma) cell line shows that ID2 is a nuclear 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 cytoplasmic distribution (HELA, SH-SY5Y) or nuclear distribution (SAOS2).