

PRODUCT INFORMATION

Product name : LMX1A antibody

Product type : Primary antibodies

Description : Rabbit polyclonal to LMX1A

Immunogen : 3 synthetic peptides (human) conjugated to KLH

Reacts with : Hu, Ms

Tested applications : ELISA, WB and IF

GENE INFORMATION

Gene Symbol : LMX1A

Gene Name : LIM homeobox transcription factor 1, alpha

Ensembl ID : ENSG00000162761

Entrez GeneID : 4009

GenBank Accession number : AY078391.1

Omim ID : 600298

Swiss-Prot : Q8TE12

Molecular weight of LMX1A : 42.747kDa (Isoform1) and 14.615kDa (Isoform LMX1A-4AB).

Function : Acts as a transcriptional activator by binding to an A/T-rich sequence, the FLAT element, in the insulin gene promoter. Required for development of the roof plate and, in turn, for specification of dorsal cell fates in the CNS and developing vertebrae

Expected subcellular localization : Nucleus.

Expected tissue specificity : Isoform 1 is expressed in many tissues. Not found in heart, liver, spleen and testis. Relatively highly expressed in fetal brain. Isoform LMX1A-4B is expressed in testis.

Summary : Insulin is produced exclusively by the beta cells in the islets of Langerhans in the pancreas. The level and beta-cell specificity of insulin gene expression are regulated by a set of nuclear genes that bind to specific sequences within the promoter of the insulin gene (INS; MIM 176730) and interact with RNA polymerase to activate or repress transcription. LMX1 is a homeodomain protein that binds an A/T-rich sequence in the insulin promoter and stimulates transcription of insulin (German et al., 1994 [PubMed 7698771]).[supplied by OMIM]

APPLICATION NOTE

Recommended dilution :

- **ELISA:** Antibody specificity was verified by direct ELISA against the 3 immunogen peptides. A titer of 1/8000 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.

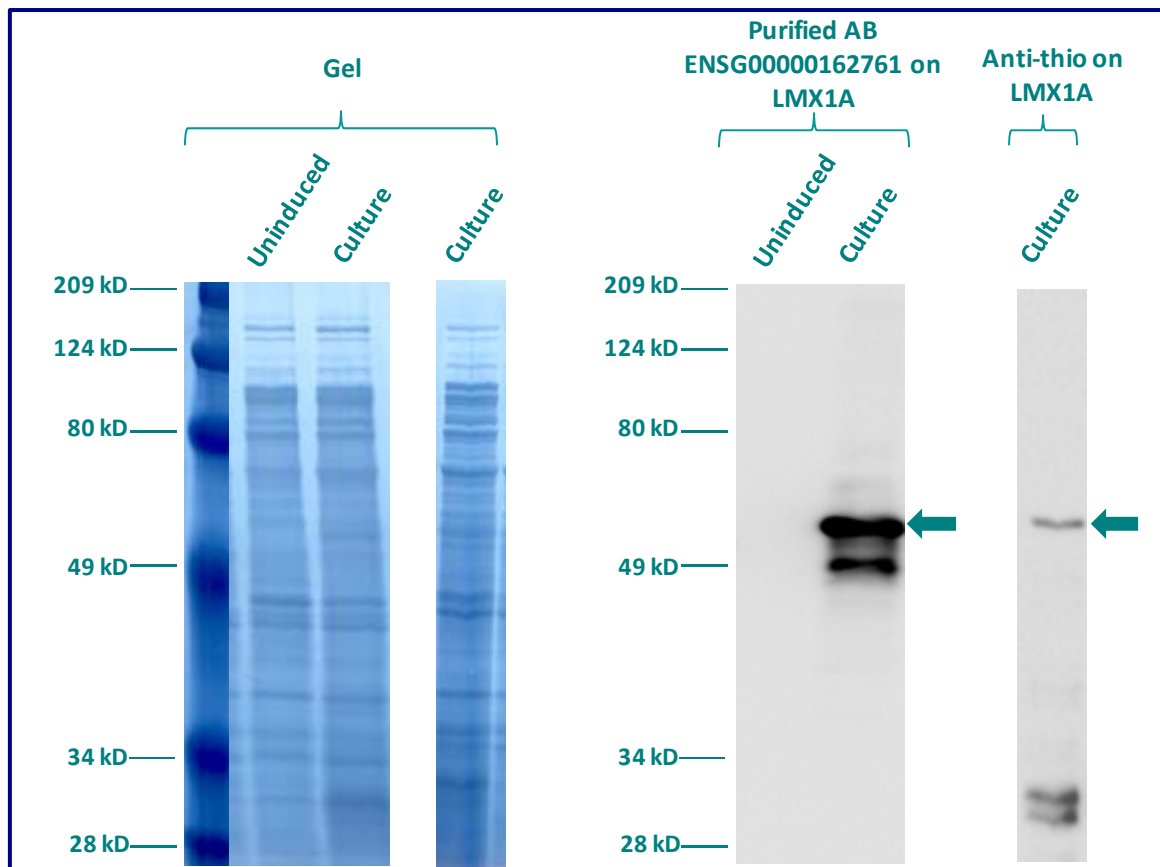
WESTERN BLOT ON RECOMBINANT PROTEIN

The purified antibody ENSG00000162761 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 LMX1A : 56.7kDa (42.7kDa + 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 ENSG00000162761 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 1/1000
- 60 minutes of incubation

IMMUNOFLUORESCENCE ANALYSIS

Immunofluorescence analysis of LIM homeobox transcription factor 1-alpha (LMX1A) expression in 5 cells lines (HELA, 293T/17, Capan-2, SH-SY5Y, Skin 3,44). The purified Antibody ENSG00000162761 has been tested at 1/5000.

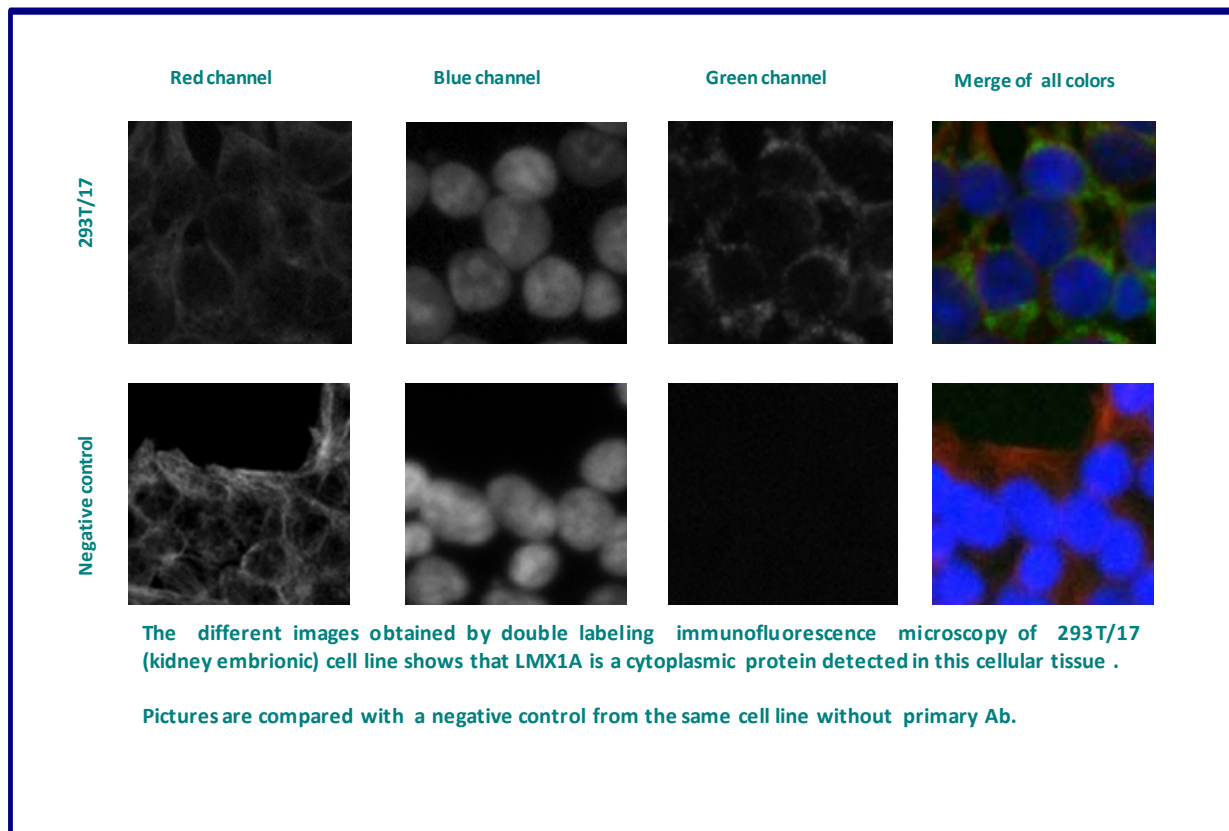
Red staining : cytoskeleton (microtubules/ α -tubuline)

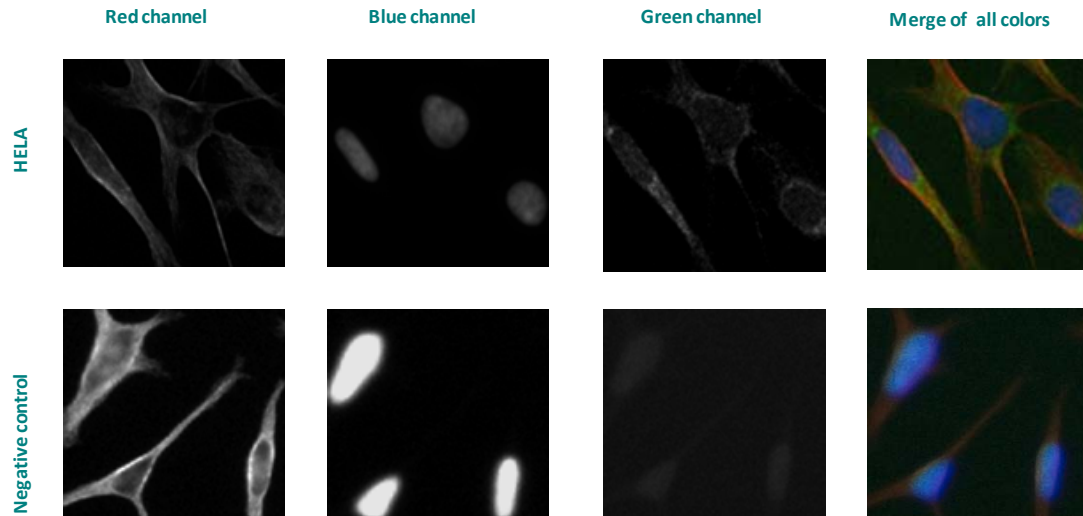
Blue staining : nucleus (Hoechst)

Green staining : anti- LMX1A antibody (purified)

Expected subcellular location : Nucleus

Expected tissue specificity : Isoform 1 is expressed in many tissues. Not found in heart, liver, spleen and testis. Relatively highly expressed in fetal brain. Isoform LMX1A-4B is expressed in testis.





The different images obtained by double labeling immunofluorescence microscopy of HELA (cervix adenocarcinoma) cell line shows that LMX1A is a 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 cytoplasmic distribution.