Anti-SALL4 Antibody [A9G9-R]

HA601168

Product Type:	Recombinant Mouse monoclonal IgG1, primary antibodies
Species reactivity:	Human
Applications:	WB, IHC-P, IF-Tissue, IF-Cell, FC
Molecular Wt:	Predicted band size: 112 kDa
Clone number:	A9G9-R

Description: Sal-like protein 4 (SALL4) is a transcription factor encoded by a member of the Spalt-like (SALL) gene family, SALL4. The SALL genes were identified based on their sequence homology to Spalt, which is a homeotic gene originally cloned in Drosophila melanogaster that is important for terminal trunk structure formation in embryogenesis and imaginal disc development in the larval stages. There are four human SALL proteins (SALL1, 2, 3, and 4) with structural homology and playing diverse roles in embryonic development, kidney function, and cancer. The SALL4 gene encodes at least three isoforms, termed A, B, and C, through alternative splicing, with the A and B forms being the most studied. SALL4 can alter gene expression changes through its interaction with many co-factors and epigenetic complexes. It is also known as a key embryonic stem cell (ESC) factor.

Immunogen: Recombinant protein within human SALL4 aa 904-1,053/1,053.

Positive control: NCCIT cell lysates, human seminoma tissue, human embryonal carcinoma tissue, human testis tissue, NCCIT.

Subcellular location: Cytoplasm, Nucleus.

Database links: SwissProt: Q9UJQ4 Human

Recommended Dilutions:

WB	1:2,000
IHC-P	1:2,000-1:4,000
IF-Tissue	1:200
IF-Cell	1:100
FC	1:500-1:1,000

Storage Buffer: PBS (pH7.4), 0.1% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

Storage Instruction: Store at +4℃ after thawing. Aliquot store at -20℃. Avoid repeated freeze / thaw cycles.

Purity: Protein A affinity purified.

Images

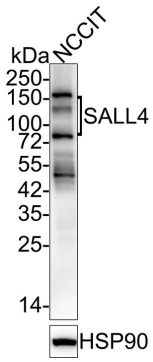

Fig1: Western blot analysis of SALL4 on NCCIT cell lysates with Mouse anti-SALL4 antibody (HA601168) at 1/2,000 dilution.

Lysates/proteins at 20 µg/Lane.

Predicted band size: 112 kDa
Observed band size: 140/110/75 kDa

Exposure time: 1 minute 26 seconds;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (HA601168) at 1/2,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Mouse IgG - HRP Secondary Antibody (HA1006) at 1/50,000 dilution was used for 1 hour at room temperature.

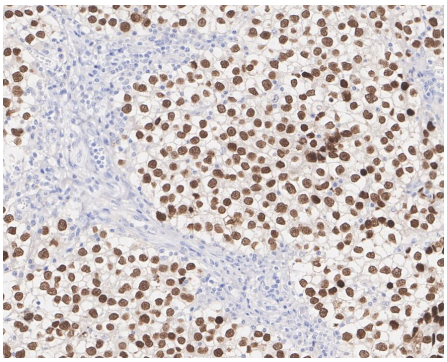

Fig2: Immunohistochemical analysis of paraffin-embedded human seminoma tissue with Mouse anti-SALL4 antibody (HA601168) at 1/4,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA601168) at 1/4,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

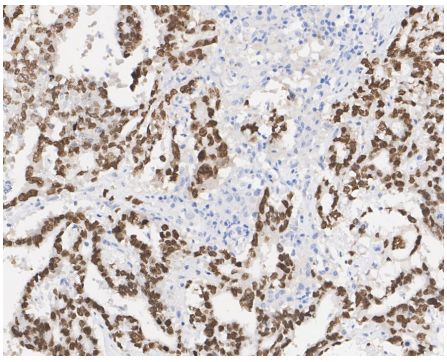

Fig3: Immunohistochemical analysis of paraffin-embedded human embryonal carcinoma tissue with Mouse anti-SALL4 antibody (HA601168) at 1/2,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA601168) at 1/2,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

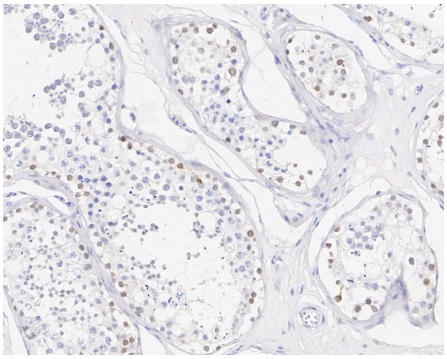

Fig4: Immunohistochemical analysis of paraffin-embedded human testis tissue with Mouse anti-SALL4 antibody (HA601168) at 1/2,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA601168) at 1/2,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

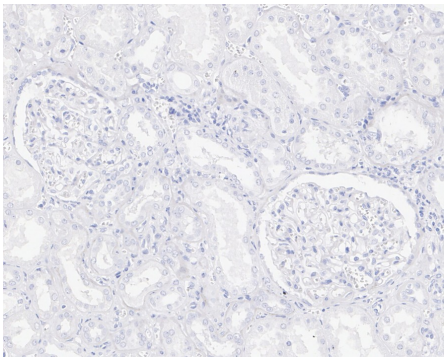

Fig5: Immunohistochemical analysis of paraffin-embedded human kidney tissue (negative control) with Mouse anti-SALL4 antibody (HA601168) at 1/4,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA601168) at 1/4,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

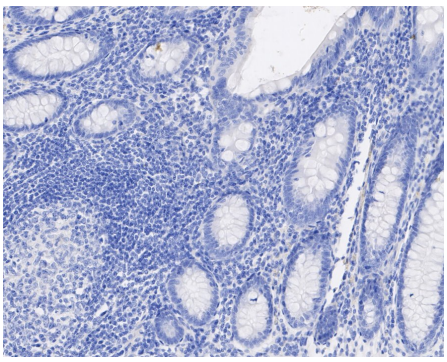

Fig6: Immunohistochemical analysis of paraffin-embedded human appendix tissue (negative control) with Mouse anti-SALL4 antibody (HA601168) at 1/4,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA601168) at 1/4,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

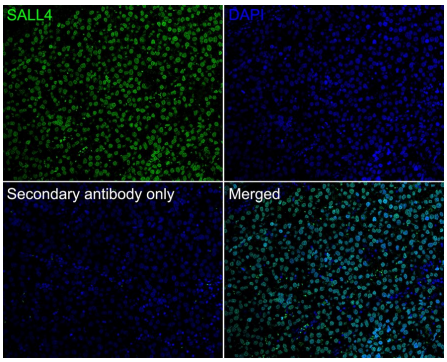

Fig7: Immunofluorescence analysis of paraffin-embedded human seminoma tissue labeling SALL4 with Mouse anti-SALL4 antibody (HA601168) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 10% negative goat serum for 1 hour at room temperature, washed with PBS, and then probed with the primary antibody (HA601168, green) at 1/200 dilution overnight at 4 ℃, washed with PBS. Goat Anti-Mouse IgG H&L (iFluor™ 488, HA1125) was used as the secondary antibody at 1/1,000 dilution. Nuclei were counterstained with DAPI (blue).

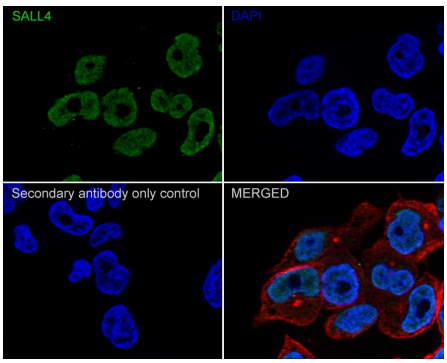

Fig8: Immunocytochemistry analysis of NCCIT cells labeling SALL4 with Mouse anti-SALL4 antibody (HA601168) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Mouse anti-SALL4 antibody (HA601168) at 1/100 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Mouse IgG H&L (iFluor™ 488, HA1125) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

beta Tubulin (ET1602-4, red) was stained at 1/100 dilution overnight at +4℃. Goat Anti-Rabbit IgG H&L (iFluor™ 594, HA1122) were used as the secondary antibody at 1/1,000 dilution.

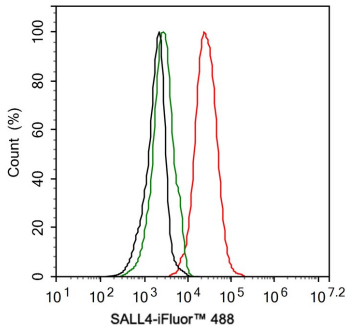

Fig9: Flow cytometric analysis of NCCIT cells labeling SALL4.

Cells were fixed and permeabilized. Then stained with the primary antibody (HA601168, 1ug/ml) (red) compared with Mouse IgG Isotype Control (green). After incubation of the primary antibody at +4℃ for an hour, the cells were stained with a iFluor™ 488 conjugate-Goat anti-Mouse IgG Secondary antibody (HA1125) at 1/1,000 dilution for 30 minutes at +4℃. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

Background References

1. Sun B et al. SALL4 Oncogenic Function in Cancers: Mechanisms and Therapeutic Relevance. Int J Mol Sci. 2022 Feb
2. Moein S et al. SALL4: An Intriguing Therapeutic Target in Cancer Treatment. Cells. 2022 Aug