Anti-SALL4 Antibody [A9G10]

HA601145



Product Type: Mouse monoclonal IgG2b, primary antibodies

Species reactivity: Human

Applications: IHC-P, WB, FC, IF-Cell

Molecular Wt: Predicted band size: 112 kDa

Clone number: A9G10

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: Human seminoma tissue, human testis tissue, NCCIT cell lysates, NCCIT.

Subcellular location: Cytoplasm, Nucleus.

Database links: SwissProt: Q9UJQ4 Human

Recommended Dilutions:

IHC-P 1:4,000
WB 1:1,000
FC 1:500-1:1,000
IF-Cell 1:3,000

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

Storage Instruction: Store at +4 $^{\circ}$ C after thawing. Aliquot store at -20 $^{\circ}$ C. Avoid repeated freeze / thaw cycles.

Purity: Protein A affinity purified.

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Images

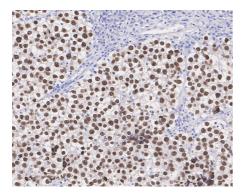


Fig1: Immunohistochemical analysis of paraffin-embedded human seminoma tissue with Mouse anti-SALL4 antibody (HA601145) at 1/4,000 dilution.

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 ddH₂O and PBS, and then probed with the primary antibody (HA601145) 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.

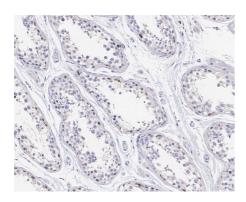


Fig2: Immunohistochemical analysis of paraffin-embedded human testis tissue with Mouse anti-SALL4 antibody (HA601145) at 1/4,000 dilution.

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 $_2$ O and PBS, and then probed with the primary antibody (HA601145) 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.

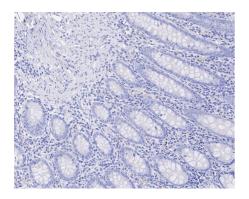


Fig3: Immunohistochemical analysis of paraffin-embedded human appendix tissue (Negative) with Mouse anti-SALL4 antibody (HA601145) at 1/4,000 dilution.

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 ddH₂O and PBS, and then probed with the primary antibody (HA601145) 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.

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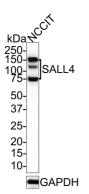


Fig4: Western blot analysis of SALL4 on NCCIT cell lysates with Mouse anti-SALL4 antibody (HA601145) at 1/1,000 dilution.

Lysates/proteins at 20 µg/Lane.

Predicted band size: 112 kDa

Observed band size: 150/110/70 kDa

Exposure time: 1 minute; ECL: K1801;

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 (HA601145) at 1/1,000 dilution was used in 5% NFDM/TBST at room temperature for 2 hours. Goat Anti-Mouse IgG - HRP Secondary Antibody (HA1006) at 1:100,000 dilution was used for 1 hour at room temperature.

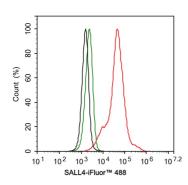


Fig5: Flow cytometric analysis of NCCIT cells labeling SALL4.

Cells were fixed and permeabilized. Then stained with the primary antibody (HA601145, 1ug/ml) (red) compared with Mouse IgG1 Isotype Control (green). After incubation of the primary antibody at +4°C 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°C. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

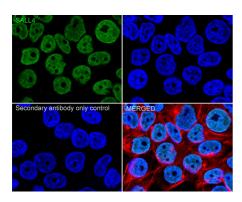


Fig6: Immunocytochemistry analysis of NCCIT cells labeling SALL4 with Mouse anti-SALL4 antibody (HA601145) at 1/3,000 dilution.

Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 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 (HA601145) at 1/3,000 dilution in 1% BSA in PBST overnight at 4 $^{\circ}\mathrm{C}$. Goat Anti-Mouse IgG H&L (iFluor $^{\dagger}\mathrm{M}$ 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°C. Goat Anti-Rabbit IgG H&L (iFluor™ 594, HA1122) were used as the secondary antibody at 1/1,000 dilution.

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Note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

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