Anti-SMYD3 Antibody [JM73-63]

ET1705-58



Product Type: Species reactivity: Applications: Molecular Wt: Clone number:	Recombinant Rabbit monoclonal IgG, primary antibodies Human, Mouse, Rat WB, IF-Cell, IF-Tissue, IHC-P, FC, IP Predicted band size: 49 kDa JM73-63
Description:	Belongs to the histone-lysine methyltransferase family. Contains 1 MYND-type zinc finger. Contains 1 SET domain. Histanes methyltransferase. Methylans 'Lys-4' of histone H3, inducing di- and tri-methylation, but not monomethylation. Plays an important role in transcriptional activation as a member of an RNA polymerase complex. Binds DNA containing 5'-CCCTCC- 3 'or 5'-GAGGGG-3'.
lmmunogen:	Synthetic peptide within Human SMYD3 aa 251-300 / 428.
Positive control:	Rat kidney tissue lysate, Mouse spleen tissue lysate, Rat spleen tissue lysate, PC-12 cell lysate, NIH-3T3 cell lysate, HepG2, Hela cell lysate, LOVO, Hela, human tonsil tissue, human colon cancer tissue, human placenta tissue, mouse stomach tissue.
Subcellular location:	Cytoplasm. Nucleus.
Database links:	SwissProt: Q9H7B4 Human Q9CWR2 Mouse
Recommended Dilutions:	
WB	1:500-1:2,000
IF-Cell	1:50-1:200
	1:50-1:200
	1:50-1:200
IP	Use at an assay dependent concentration.
Storage Buffer:	1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.
Storage Instruction:	Store at +4 $^\circ\!\!C$ after thawing. Aliquot store at -20 $^\circ\!\!C$ or -80 $^\circ\!\!C$. Avoid repeated freeze / thaw cycles.
Purity:	Protein A affinity purified.

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Images



Fig1: Western blot analysis of SMYD3 on different lysates with Rabbit anti-SMYD3 antibody (ET1705-58) at 1/1000 dilution.

Lane 1: Rat kidney tissue lysate Lane 2: PC-12 cell lysate Lane 3: Hela cell lysate Lane 4: Mouse spleen tissue lysate

Lane 5: NIH-3T3 cell lysate

Lane 6: Rat spleen tissue lysate

Lysates/proteins at 10 µg/Lane.

Predicted band size: 39 kDa Observed band size: 39 kDa

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (ET1705-58) at 1/1000 dilution was used in 5% NFDM/TBST at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:300,000 dilution was used for 1 hour at room temperature.



Fig2: "Immunocytochemistry analysis of Hela cells labeling SMYD3 with Rabbit anti-SMYD3 antibody (ET1705-58) at 1/50 dilution.

Cells were fixed in 4% paraformaldehyde for 10 minutes at 37 $^{\circ}$ C, permeabilized with 0.05% Triton X-100 in PBS for 20 minutes, and then blocked with 2% negative goat serum for 30 minutes at room temperature. Cells were then incubated with Rabbit anti-SMYD3 antibody (ET1705-58) at 1/50 dilution in 2% negative goat serum overnight at 4 $^{\circ}$ C. Goat Anti-Rabbit IgG H&L (iFluorTM 488, HA1121) 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."

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Fig3: Immunocytochemistry analysis of HepG2 cells labeling SMYD3 with Rabbit anti-SMYD3 antibody (ET1705-58) at 1/200 dilution.

Cells were fixed in 4% paraformaldehyde for 10 minutes at 37 $^{\circ}$ C, permeabilized with 0.05% Triton X-100 in PBS for 20 minutes, and then blocked with 2% negative goat serum for 30 minutes at room temperature. Cells were then incubated with Rabbit anti-SMYD3 antibody (ET1705-58) at 1/200 dilution in 2% negative goat serum overnight at 4 $^{\circ}$ C. Goat Anti-Rabbit IgG H&L (iFluor M 488, HA1121) 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.



Fig4: Immunocytochemistry analysis of LOVO cells labeling SMYD3 with Rabbit anti-SMYD3 antibody (ET1705-58) at 1/200 dilution.

Cells were fixed in 4% paraformaldehyde for 10 minutes at 37 $^{\circ}$ C, permeabilized with 0.05% Triton X-100 in PBS for 20 minutes, and then blocked with 2% negative goat serum for 30 minutes at room temperature. Cells were then incubated with Rabbit anti-SMYD3 antibody (ET1705-58) at 1/200 dilution in 2% negative goat serum overnight at 4 $^{\circ}$ C. Goat Anti-Rabbit IgG H&L (iFluor M 488, HA1121) 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..



Fig5: Immunohistochemical analysis of paraffin-embedded human tonsil tissue using anti-SMYD3 antibody. Counter stained with hematoxylin.

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Fig6: Immunohistochemical analysis of paraffin-embedded human colon cancer tissue with Rabbit anti-SMYD3 antibody (ET1705-58) at 1/50 dilution.Hela cells

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 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 (ET1705-58) at 1/50 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: Immunohistochemical analysis of paraffin-embedded human placenta tissue with Rabbit anti-SMYD3 antibody (ET1705-58) at 1/50 dilution.Hela cells

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 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 (ET1705-58) at 1/50 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.



Fig8: Immunohistochemical analysis of paraffin-embedded mouse stomach tissue with Rabbit anti-SMYD3 antibody (ET1705-58) at 1/50 dilution.Hela cells

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 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 (ET1705-58) at 1/50 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.



Fig9: Flow cytometric analysis of Hela cells with SMYD3 antibody at 1/100 dilution (red) compared with an unlabelled control (cells without incubation with primary antibody; black).

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

Background References

- 1. Liu T et al. Master redox regulator Trx1 upregulates SMYD1 & modulates lysine methylation. Biochim Biophys Acta 1854:1816-22 (2015).
- 2. Wang HF et al. Associations of the variable number of tandem repeats polymorphism in the SMYD3 gene with risk and prognosis of esophageal cancer: a case-control study. Neoplasma. doi: 10.4149/neo_2017_613 (2017).

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Applications:WB=Western blot IHC-P=Immunohistochemistry (paraffin) IF-Cell=Immunofluorescence (Cell) IF-Tissue=Immunofluorescence (Tissue) FC=Flow cytometry IP=Immunoprecipitation

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