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EZH2 Antibody

Affinity Purified Rabbit Polyclonal Antibody (Pab)
Catalog # AP2512d

Product Information

Application	WB, IHC-P, IF, FC, E
Primary Accession	Q15910
Reactivity	Human, Mouse
Host	Rabbit
Clonality	Polyclonal
Isotype	Rabbit IgG
Clone Names	RB23144
Calculated MW	85363
Antigen Region	1-296

Additional Information

Gene ID	2146
Other Names	Histone-lysine N-methyltransferase EZH2, ENX-1, Enhancer of zeste homolog 2, Lysine N-methyltransferase 6, EZH2, KMT6
Target/Specificity	This EZH2 antibody is generated from rabbits immunized with a recombinant fragment (N-term) protein from human EZH2.
Dilution	WB~~1:1000 IHC-P~~1:100~500 IF~~1:10~50 FC~~1:10~50 E~~Use at an assay dependent concentration.
Format	Purified polyclonal antibody supplied in PBS with 0.05% (V/V) Proclin 300. This antibody is purified through a protein A column, followed by peptide affinity purification.
Storage	Maintain refrigerated at 2-8°C for up to 2 weeks. For long term storage store at -20°C in small aliquots to prevent freeze-thaw cycles.
Precautions	EZH2 Antibody is for research use only and not for use in diagnostic or therapeutic procedures.

Protein Information

Name	EZH2 (HGNC:3527)
Synonyms	KMT6
Function	Polycomb group (PcG) protein. Catalytic subunit of the PRC2/EED-EZH2 complex, which methylates 'Lys-9' (H3K9me) and 'Lys-27' (H3K27me) of

histone H3, leading to transcriptional repression of the affected target gene. Able to mono-, di- and trimethylate 'Lys-27' of histone H3 to form H3K27me1, H3K27me2 and H3K27me3, respectively. Displays a preference for substrates with less methylation, loses activity when progressively more methyl groups are incorporated into H3K27, H3K27me0 > H3K27me1 > H3K27me2 (PubMed:22323599, PubMed:30923826). Compared to EZH1-containing complexes, it is more abundant in embryonic stem cells and plays a major role in forming H3K27me3, which is required for embryonic stem cell identity and proper differentiation. The PRC2/EED-EZH2 complex may also serve as a recruiting platform for DNA methyltransferases, thereby linking two epigenetic repression systems. Genes repressed by the PRC2/EED-EZH2 complex include HOXC8, HOXA9, MYT1, CDKN2A and retinoic acid target genes. EZH2 can also methylate non-histone proteins such as the transcription factor GATA4 and the nuclear receptor RORA. Regulates the circadian clock via histone methylation at the promoter of the circadian genes. Essential for the CRY1/2-mediated repression of the transcriptional activation of PER1/2 by the CLOCK-BMAL1 heterodimer; involved in the di and trimethylation of 'Lys-27' of histone H3 on PER1/2 promoters which is necessary for the CRY1/2 proteins to inhibit transcription.

Cellular Location

Nucleus. Note=Localizes to the inactive X chromosome in trophoblast stem cells. {ECO:0000250|UniProtKB:Q61188}

Tissue Location

In the ovary, expressed in primordial follicles and oocytes and also in external follicle cells (at protein level) (PubMed:31451685). Expressed in many tissues (PubMed:14532106) Overexpressed in numerous tumor types including carcinomas of the breast, colon, larynx, lymphoma and testis (PubMed:14532106)

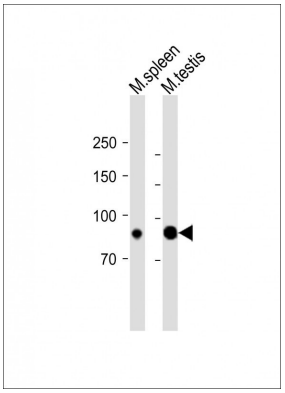
Background

EZH2 is a protein that transfers sulfate to the C-4 hydroxyl of beta-1,4-linked GalNAc flanked by GlcUA residues in chondroitin.

References

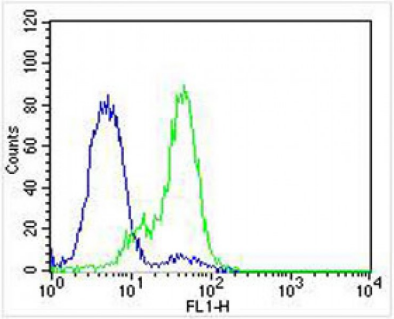
Kang,H.G., et.al., J. Biol. Chem. 277 (38), 34766-34772 (2002)
Hiraoka,N., et.al., J. Biol. Chem. 275 (26), 20188-20196 (2000)

Images

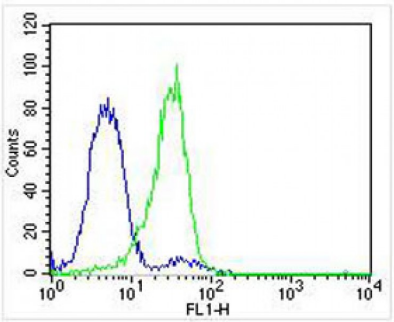


All lanes: Anti-EZH2 Antibody at 1:2000 dilution Lane 1: Mouse spleen lysate Lane 2: Mouse testis lysate Lysates/proteins at 20 µg per lane. Secondary: Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated (ASP1615) at 1/15000 dilution. Observed band size: 85 KDa Blocking/Dilution buffer: 5% NFDM/TBST.

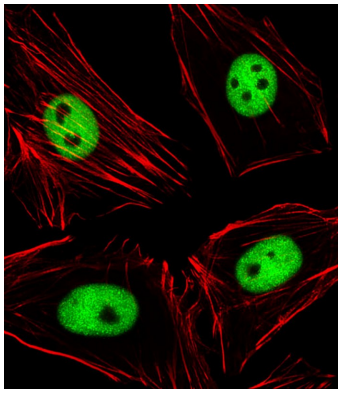
Overlay histogram showing Hela cells stained with AP2512D (green line). The cells were fixed with 2%



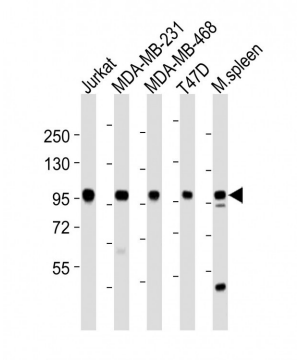
paraformaldehyde (10 min) and then permeabilized with 90% methanol for 10 min. The cells were then icubated in 2% bovine serum albumin to block non-specific protein-protein interactions followed by the antibody (AP2512D, 1:25 dilution) for 60 min at 37ºC. The secondary antibody used was Goat-Anti-Rabbit IgG, DyLight® 488 Conjugated Highly Cross-Adsorbed(NA168821) at 1/400 dilution for 40 min at 37ºC. Isotype control antibody (blue line) was rabbit IgG (1µg/1x10^6 cells) used under the same conditions. Acquisition of >10, 000 events was performed.



Overlay histogram showing Hela cells stained with AP2512D (green line). The cells were fixed with 2% paraformaldehyde (10 min) and then permeabilized with 90% methanol for 10 min. The cells were then icubated in 2% bovine serum albumin to block non-specific protein-protein interactions followed by the antibody (AP2512D, 1:25 dilution) for 60 min at 37ºC. The secondary antibody used was Goat-Anti-Rabbit IgG, DyLight® 488 Conjugated Highly Cross-Adsorbed(NA168821) at 1/400 dilution for 40 min at 37ºC. Isotype control antibody (blue line) was rabbit IgG (1µg/1x10^6 cells) used under the same conditions. Acquisition of >10, 000 events was performed.

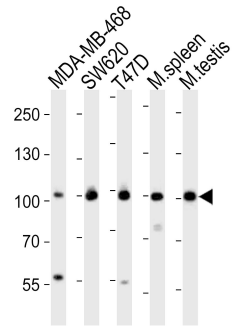


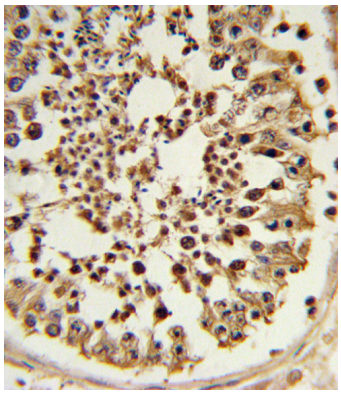
Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0. 1% Triton X-100 permeabilized Hela (Human cervical epithelial adenocarcinoma cell line) cells labeling EZH2 with AP2512d at 1/25 dilution, followed by Alexa Fluor 488-conjugated goat anti-rabbit IgG (1583138) secondary antibody at 1/400 dilution (green). Confocal image showing nuclear staining on Hela cell line. Cytoplasmic actin is detected with Alexa Fluor® 555 conjugated with Phalloidin (OB16636430) at 1/100 dilution (red).



All lanes : Anti-EZH2 Antibody at 1:2000 dilution Lane 1: Jurkat whole cell lysates Lane 2: MDA-MB-231 whole cell lysates Lane 3: MDA-MB-468 whole cell lysates Lane 4: T47D whole cell lysates Lane 5: mouse spleen lysates Lysates/proteins at 20 µg per lane. Secondary Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/10000 dilution Predicted band size : 85 kDa Blocking/Dilution buffer: 5% NFDM/TBST.

Western blot analysis of lysates from MDA-MB-468, SW620, T47D cell line, mouse spleen, mouse testis tissue (from left to right), using EZH2 Antibody(Cat. #AP2512d). AP2512d was diluted at 1:1000 at each lane. A goat anti-rabbit IgG H&L(HRP) at 1:10000 dilution was used as the secondary antibody. Lysates at 20ug per lane.





Formalin-fixed and paraffin-embedded human testis tissue reacted with EZH2 Antibody, which was peroxidase-conjugated to the secondary antibody, followed by DAB staining. This data demonstrates the use of this antibody for immunohistochemistry; clinical relevance has not been evaluated.

Citations

- Link between the EZH2 noncanonical pathway and microtubule organization center polarization during early T lymphopoiesis
- Role of epigenetic regulation on the induction of apoptosis in Jurkat leukemia cells by white grape pomace rich in phenolic compounds.
- Clinicopathological significance of EZH2 mRNA expression in patients with hepatocellular carcinoma.