diagenode
A Hologic Company

TECHNICAL DATASHEET

H3K4me3T6p polyclonal antibody - Classic

Cat. No. C15410281
Type: Polyclonal
Source: Rabbit
Lot #: 001
Size: 50 µg
Concentration: 0.55 µg/µl

Specificity: Human, mouse, C. elegans, rat, chicken, Xenopus, Drosophila, plant
Purity: Affinity purified
Storage: Store at -20°C; for long storage, store at -80°C. Avoid multiple freeze-thaw cycles.
Precautions: This product is for research use only. Not for use in diagnostic or therapeutic procedures.

Applications

	Suggested dilution	Results
ChIP	2-5 µg/million cells	Figure 1
Immunohistochemistry	1:50	
IF	1:50	Figure 2
Western blot	1:500	Figure 3, 4
Dot blot	1:1,000	Figure 5

Target description

Chromatin is the arrangement of DNA and proteins in which chromosomes are formed. Correspondingly, chromatin is formed from nucleosomes, which are comprised of a set of four histone proteins (H2A, H2B, H3, H4) wrapped with DNA. Chromatin is a very dynamic structure in which numerous post-translational modifications work together to activate or repress the availability of DNA to be copied, transcribed, or repaired. These marks decide which DNA will be open and commonly active (euchromatin) or tightly wound to prevent access and activation (heterochromatin). Common histone modifications include methylation of lysine and arginine, acetylation of lysine, phosphorylation of threonine and serine, and sumoylation, biotinylation, and ubiquitylation of lysine. In particular, trimethylation of lysine 4 on H3 (H3K4Me3) is a well known mark of gene activation. However, the role of phosphorylation at threonine 6 on H3 (H3T6p) is more obscure. Yet recently, the two modifications have been shown to interact with each other. When H3T6 is phosphorylated by protein kinase C beta 1 (PRKCbeta), the histone demethylase LSD1 is prevented from removing methyl groups from H3K4. This same study also correlated high levels of T6p and PRKCbeta as a possible marker for prostate cancer, as well as tumor progression in xenografts.

Results

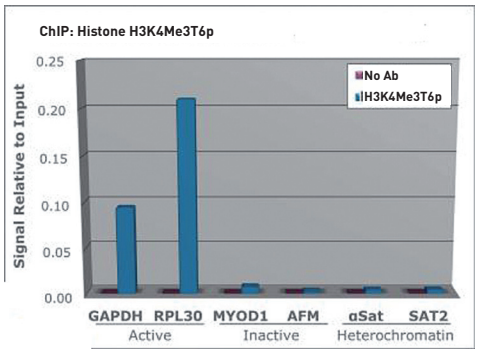

Figure 1. H3K4me3T6p antibody ChIP results

Chromatin Immunoprecipitation of H3K4me3T6p antibody. Chromatin from one million formaldehyde cross-linked Hela cells was used with 2 µg of H3K4me3T6p antibody alongside a no antibody (No Ab) control, DNA was measured by qPCR and normalized to total input.

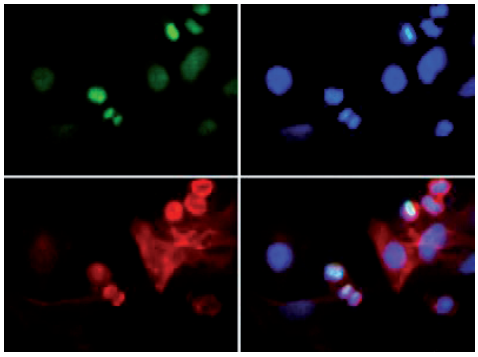

Figure 2. H3K4me3T6p antibody Immunofluorescence results

Immunofluorescence of H3K4me3T6p antibody. Tissue: HeLa cells. Fixation: 0.5% PFA. Primary antibody used at a 1:50 dilution for 1 h at RT. Secondary antibody: FITC secondary antibody at 1:10,000 for 45 min at RT. Localization: Histone H3K4me3T6p is nuclear and chromosomal. Staining: H3K4me3T6p is expressed in green and the nuclei and alpha-tubulin are counterstained with DAPI (blue) and Dylight 594 (red).

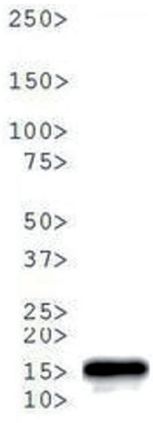

Figure 3. H3K4me3T6p antibody Western blot results

Western Blot of Rabbit H3K4me3T6p antibody. 30 µg C. elegans embryo lysate. Primary antibody at 1:500 overnight at 4°C. Secondary antibody: IRDye800™ rabbit secondary antibody at 1:10,000 for 45 min at RT. Predicted/Observed size: ~15 kDa. Other band(s): None.

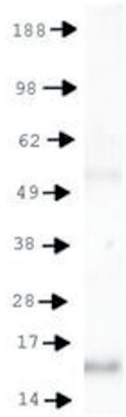

Figure 4. H3K4me3T6p antibody Western blot results

Western Blot of H3K4me3T6p antibody. 30 µg HeLa histone extracts. Primary antibody at 1:500 overnight at 4°C. Secondary antibody: IRDye800™ rabbit secondary antibody at 1:10,000 for 45 min at RT. Predicted/Observed size: ~15 kDa. Other band(s): None.

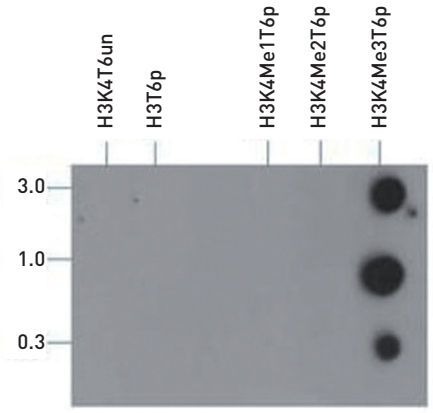

Figure 5. H3K4me3T6p antibody Dot blot results

Dot Blot of Rabbit H3K4me3T6p antibody. Lane 1: Unmodified. Lane 2: T6p. Lane 3: K4Me1T6p. Lane 4: K4Me2T6p. Lane 5: K4Me3T6p. Load: 3, 1, and 0.3 picomoles of peptide. Primary antibody used at a 1:1,000 dilution for 45 min at 4°C. Secondary antibody: Dylight™488 rabbit secondary antibody at 1:10,000 for 45 min at RT.

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