

### TECHNICAL DATASHEET

# H3K27me3S28p polyclonal antibody - Classic

Cat. No. C15410293
Type: Polyclonal
Source: Rabbit
Lot #: 001

**Size:** 50 μg

Concentration: 0.66 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
IF	1:200	Figure 2, 3
Western blot	1:500	Figure 4
Immunochemistry	1:50	
Dot blot	1:500 - 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. Specifically, trimethylation of K27 is associated with gene silencing, whereas pS28 is associated with mitotis and immediate early genes.



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#### Results

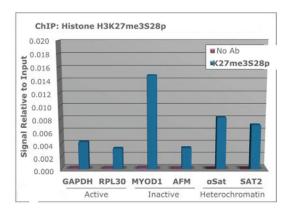


Figure 1. ChIP

Chromatin Immunoprecipitation using the H3K27me3S28p antibody. Chromatin from one million formaldehyde cross-linked HeLa cells was used with 2  $\mu g$  of H3K27me3S28p and 20  $\mu l$  of magnetic IgG beads per immunoprecipitation. A no antibody [No Ab] control was also used. Immunoprecipitated DNA was quantified using quantitative PCR and normalized to the input chromatin.

Figure 2. Immunofluorescence

Immunofluorescence of H3K27me3S28p antibody. Tissue: HeLa cells. Fixation: 0.5% PFA. Primary antibody used at a 1:200 dilution for 1 h at RT. Secondary antibody: Dylight 488 secondary antibody at 1:10,000 for 45 min at RT. Localization: Histone H3K27me3S28p is nuclear and chromosomal. Staining: Histone H3K27me3S28p is expressed in green, nuclei and alpha-tubulin are counterstained with DAPI (blue) and Dylight 550 (red).



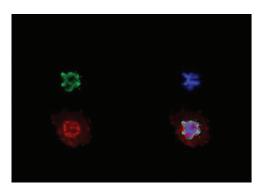
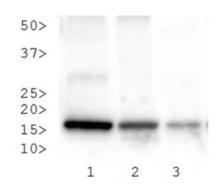


Figure 3. Immunofluorescence

Immunofluorescence of H3K27me3S28p antibody. Tissue: HeLa cells. Fixation: 0.5% PFA. Primary antibody used at a 1:200 dilution for 1 h at RT. Secondary antibody: Dylight 488 secondary antibody at 1:10,000 for 45 min at RT. Localization: Histone H3K27me3S28p is nuclear and chromosomal. Staining: Histone H3K27me3S28p is expressed in green, nuclei and alpha-tubulin are counterstained with DAPI (blue) and Dylight 550 (red).

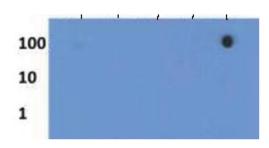
Figure 4. Western Blot

Western Blot of H3K27me3S28p antibody. Lane 1: HeLa histone extracts. Lane 2: NIH-3T3 histone extracts. Lane 3: C. elegans embryo cell lysate. Load: 30  $\mu$ g per lane. Primary antibody used at 1  $\mu$ g/ml overnight at 4°C. Secondary antibody: IRDye800<sup>TM</sup> rabbit secondary antibody at 1:10,000 for 45 min at RT. Predicted/Observed size: ~15 kDa. Other band(s): None.





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#### Figure 5. Dot Blot

Dot Blot of H3K27me3S28p antibody. Lane 1: S28p/K27 unmodified. Lane 2: S28p N-Term. Lane 3: S28p C-term. Lane 4: K27Me3. Lane 5: S28p/K27Me3. Load: 1, 10, and 100 picomoles of peptide. Primary antibody at 1  $\mu$ g/ml for 45 min at 4°C. Secondary antibody: Dylight  $^{TM}$ 488 rabbit secondary antibody at 1:10,000 for 45 min at RT.

LIEGE SCIENCE PARK
Rue Bois Saint-Jean, 3
4102 Seraing (Ougrée) - Belgium
Tel: +32 4 364 20 50
Fax: +32 4 364 20 51
orders@diagenode.com
info@diagenode.com

400 Morris Avenue, Suite 101 Denville, NJ 07834 - USA Tel: +1 862 209-4680 Fax: +1 862 209-4681 orders.na@diagenode.com info.na@diagenode.com