

# Histone H3S10phS28ph antibody (pAb)

Catalog Nos: 39147, 39148

RRID: AB\_2793167 Isotype: Serum

Application(s): ChIP, ICC, IF, WB

Reactivity: Human, Wide Range Predicted

Volumes: 200 μl, 10 μl Purification: None Host: Rabbit

Molecular Weight: 17 kDa

**Background:** Histone H3 is one of the core components of the nucleosome. The nucleosome is the smallest subunit of chromatin and consists of 147 base pairs of DNA wrapped around an octamer of core histone proteins (two each of Histone H2A, Histone H2B, Histone H3 and Histone H4). Chromatin is subject to a variety of chemical modifications, including post-translational modifications of the histone proteins and the methylation of cytosine residues in the DNA. Reported histone modifications include acetylation, methylation, phosphorylation, ubiquitylation, glycosylation, ADP-ribosylation, carbonylation and SUMOylation; these modifications play a major role in regulating gene expression.

Ser10 phosphorylation and Ser28 phosphorylation in the tail of H3 have very similar kinetics. Both phosphorylations occur early in mitosis when chromosomes begin to condense, and during premature chromosome condensation induced in S-phase cells. These phosphorylated serines are excellent mitotic markers.

**Immunogen:** This Histone H3 phospho Ser10,28 antibody was raised against a peptide including phosphorylated serine 28 of human Histone H3.

**Buffer:** Rabbit serum containing 30% glycerol and 0.035% sodium azide. Sodium azide is highly toxic.

## **Application Notes:**

Applications Validated by Active Motif:

ChIP: 10 µl per ChIP

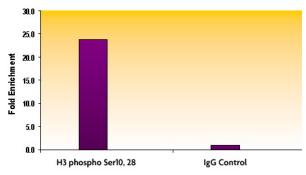
ICC/IF: 1:500 - 1:2,000 dilution WB\*: 1:1,000 - 1:10,000 dilution

\*Note: many chromatin-bound proteins are not soluble in a low salt nuclear extract and fractionate to the pellet. Therefore, we recommend a High Salt / Sonication Protocol when preparing nuclear extracts for Western blot.

**Storage and Guarantee:** Some products may be shipped at room temperature. This will not affect their stability or performance. Avoid repeated freeze/thaw cycles by aliquoting items into single-use fractions for storage at -20°C for up to 2 years. Keep all reagents on ice when not in storage. This product is guaranteed for 12 months from date of receipt.

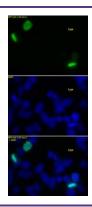
This product is for research use only and is not for use in diagnostic procedures.





## Histone H3 phospho Ser10,28 antibody tested by ChIP analysis.

Chromatin IP performed using the ChIP-IT® Express Kit (Catalog No. 53008) and HeLa Chromatin (1.5 x  $10^6$  cell equivalents per ChIP) using 10  $\mu$ I of Histone H3 phospho Ser10, 28 antibody or the equivalent amount of rabbit IgG as a negative control. Real time, quantitative PCR (RT-qPCR) was performed on DNA purified from each of the ChIP reactions using a primer pair specific for the GAPDH gene promoter. Data are presented as Fold Enrichment of the ChIP antibody signal versus the negative control IgG using the ddCT method.



#### Histone H3 phospho Ser10,28 antibody tested by immunofluorescence.

HeLa cells stained with Histone H3 phospho Ser10,28 antibody at a 1:1,000 dilution.

Top panel: Histone H3 phospho Ser10,28 antibody.

Middle: DAPI.

Bottom: Merge of both images.



#### Histone H3 phospho Ser10,28 antibody tested by Western blot.

HeLa acid extract probed with Catalog No. 39147 (1:5,000 dilution).

Lane 1: No treatment.

Lane 2: Cells treated with colcemid.