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# **ProductInformation**

Anti-Phospho-Histone H3 [pSer<sup>10</sup>]
Developed in Rabbit
IgG Fraction of Antiserum

Product Number H 0412

**Product Description** 

Anti-Phospho-Histone H3 [pSer<sup>10</sup>] is developed in rabbit using a synthetic, phosphorylated peptide [pSer<sup>10</sup>] histone H3 (7-20) corresponding to the N-terminus of human histone H3, conjugated to KLH as immunogen. This histone H3 sequence is identical in many species including mouse, rat, bovine, chicken, frog, drosophila and *C. elegans*, and is highly conserved (single amino acid substitution) in tetrahymena histone H3. Whole antiserum is fractionated and then further purified by ion-exchange chromatography. The resulting IgG fraction is further purified by absorption on the non-phosphorylated histone H3 peptide (human, amino acids 7-20).

Anti-Phospho-Histone H3 [pSer<sup>10</sup>] recognizes histone H3 phosphorylated on Ser<sup>10</sup> (17 kDa). Applications include the detection of pSer<sup>10</sup>- histone H3 by immunoblotting and by immunofluorescence. Staining of pSer<sup>10</sup>- histone H3 in immunoblotting is specifically inhibited with the [pSer<sup>10</sup>] histone H3 immunizing peptide (human, amino acids 7-20). No inhibition is observed with the respective non-phosphorylated histone H3 peptide histone H3 (human, amino acids 7-20).

Histone proteins H3, H4, H2A, H2B and H1 function as building blocks to package eukaryotic DNA into repeating nucleosome units that are folded in higherorder chromatin fibers. The relatively unstructured and highly charged N-terminal tail domains of histones, are central to the processes that modulate chromatin structure. A diverse and elaborate array of posttranslational modifications including acetylation, phosphorylation, methylation, ubiquitination and ADPribosylation occur on the N-terminal tail domains of histones.<sup>2-4</sup> Phosphorylation of H3, referred to as the nucleosomal response, is localized to a small fraction of highly acetylated H3 and occurs primarily in response to mitogenic and stress stimuli. 2, 3, 5-8 Histone H3 is phosphorylated during mitosis on at least two serine residues, Ser<sup>10</sup> and Ser<sup>28</sup>. Phosphorylation of Ser<sup>10</sup> is considered a crucial event for the onset of mitosis. It is tightly correlated with chromosome condensation during both mitosis and meiosis.

Members of the Aurora/AIK family of protein kinases are involved in the mitotic phosphorylation of histone H3 at Ser<sup>10</sup> residue. <sup>9, 10</sup> Expression of Aurora-A and Aurora-B is tightly coordinated with histone H3 phosphorylation at G2/M transition and both kinases efficiently phosphorylate H3 at Ser<sup>10</sup> *in vitro* and *in vivo*. Phosphorylation of H3 at Ser<sup>10</sup> is also directly correlated with the induction of immediate-early genes such as *c-jun*, *c-fos* and *c-myc*. PKA, Rsk-2 and Msk-1 are necessary for the histone H3 phosphorylation. <sup>11-13</sup> Mutations in Rsk-2, associated with Coffin-Lowry syndrome (CLS) in humans, or deletion of Rsk-2 in knockout mice, both result in impaired transcriptional activation of *c-fos* and a loss of EGF-induced phosphorylation of H3 *in vivo*. <sup>11, 12</sup> Msk-1 is activated by both ERK and p38 pathways, suggesting that both pathways can activate this kinase to phosphorylate histone H3. <sup>13</sup>

### Reagent

Anti-Phospho-Histone H3 [pSer<sup>10</sup>] is supplied as a solution in 0.01 M phosphate buffered saline, pH 7.4, containing 15 mM sodium azide.

## **Precautions and Disclaimer**

Due to the sodium azide content, a material safety data sheet (MSDS) for this product has been sent to the attention of the safety officer of your institution. Consult the MSDS for information regarding hazards and safe handling practices.

## Storage/Stability

For continuous use, store at 2-8 °C for up to one month. For prolonged storage, freeze in working aliquots at -20 °C. Repeated freezing and thawing is not recommended. Storage in frost-free freezers is also not recommended. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use. Working dilutions should be discarded if not used within 12 hours.

#### **Product Profile**

For immunoblotting, a minimum working antibody dilution of 1:2,000 is recommended using a whole cell extract of human acute T cell leukemia Jurkat cell line treated with nocodazole.

For indirect immunofluorescence, a minimum working antibody dilution of 1:500 is recommended using the human epitheloid carcinoma HeLa cell line treated with nocodazole.

Note: In order to obtain the best results using various techniques and preparations, we recommend determining the optimal working dilutions by titration.

#### References

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