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# **Product Information**

Monoclonal Anti- phospho-RNA polymerase II CTD (pSer⁵), Clone 1H4B6 produced in rat, purified from hybridoma cell culture

Catalog Number SAB4200638

# **Product Description**

Monoclonal Anti-phospho-RNA polymerase II CTD (pSer<sup>5</sup>) (rat IgG2b isotype) is derived from the hybridoma 1H4B6 produced by the fusion of mouse myeloma cells and lymph node cells from rat immunized with Synthetic peptide corresponding to pSer<sup>5</sup> of human RNA Polymerase II CTD repeat (GeneID: 5430). The antibody is purified from culture supernatant of hybridoma cells grown in a bioreactor.

Monoclonal Anti-phospho-RNA polymerase II CTD (pSer<sup>5</sup>) recognizes specifically phosphor-Ser<sup>5</sup> RNAPII but not phosphor-Ser<sup>2</sup> or phospho-Ser<sup>7</sup>. The specificity is found in human, monkey, bovine, canine, chicken, hamster, rat and mouse as well as in a wide variety of tissues. The product may be used in several immunochemical techniques including immunoblotting (~250 kDa) and immunofluorescence.

RNAPII also known as POLR2A is the largest subunit of RNA polymerase II, the polymerase responsible for synthesizing messenger RNA in eukaryotes. This polymerase contains a carboxy terminal domain (CTD) composed of heptapeptide repeats that are essential for polymerase activity. These repeats contain serine and threonine residues that are phosphorylated in actively transcribing RNA polymerase. The CTD functions to help couple transcription and processing of the nascent RNA and also plays roles in transcription elongation and termination. In addition, this subunit, in combination with several other polymerase subunits. forms the DNA binding domain of the polymerase, a groove in which the DNA template is transcribed into RNA.<sup>1-2</sup> CTD is being dynamically modified during transcription cycle. Phosphorylation levels on Ser<sup>5</sup> are enriched at the promoter and decrease successively towards the 3' end of genes. Cdk7 of TFIIH phosphorylates Ser<sup>5</sup> and Ser<sup>7</sup> of the CTD early in the transcription cycle in a Mediator dependent manner, which leads to the dissociation of Mediator. The recruitment of the capping machinery is the main function of phosphor-Ser<sup>5</sup>. However, other protein interactions require the phosphor-Ser<sup>5</sup> as well. 1,4

# Reagent

Supplied as a solution in 0.01 M phosphate buffered saline, pH 7.4, containing 15 mM sodium azide as a preservative.

Antibody Concentration: ~ 1.0 mg/mL

#### **Precautions and Disclaimer**

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

# Storage/Stability

For extended storage, freeze at -20  $^{\circ}$ C in working aliquots. Repeated freezing and thawing or storage in "frost-free" freezers is not recommended. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use. Working dilution samples should be discarded if not used within 12 hours.

## **Product Profile**

 $\underline{Immunoblotting} \hbox{: a working concentration of 0.5-1} \\ \mu g/mL \hbox{ is recommended using whole extracts of HeLa cells.}$ 

 $\underline{Immunofluorescence} : a working concentration of 4-8 \ \mu g/mL is recommended using HeLa cells.$ 

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

### References

- Odawara J., et al., BMC Genomics, 12, 516 (2011).
- 2. Hsin, J.P., and Manley, J.L., *Genes Dev.,* **26**, 2119-2137 (2012).
- 3. Heidemann, M., et al., *Biochim. Biophys. Acta.*, **829**, 55–62 (2013).

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