

## AbFlex<sup>®</sup> RNA Pol II antibody (rAb)

Catalog Nos: 91151, 91152

RRID: AB\_2793789 Isotype: IgG2a Application(s): ChIP, ChIP-Seq, IF, WB Reactivity: Human Quantities: 100 µg, 10 µg Purification: Protein A Host: Mouse Concentration: 1 µg/µl Molecular Weight: 217 kDa

**Background:** AbFlex<sup>®</sup> antibodies are recombinant antibodies (rAbs) that have been generated using defined DNA sequences to produce highly specific, reproducible antibodies. Each AbFlex antibody contains a 6xHis Tag, a Biotinylation Tag for enzymatic biotin conjugation using the biotin ligase, BirA, and a sortase recognition motif (LPXTG) to attach a variety of labels directly to the antibody including fluorophores, enzymatic substrates (HRP, AP), peptides, drugs as well as solid supports.

AbFlex<sup>®</sup> RNA Pol II antibody was expressed as full-length IgG with mouse immunoglobulin heavy and light chains (IgG2a isotype) in mammalian 293 cells.

RNA pol II (RNA polymerase II) is responsible for synthesizing messenger RNA in eukaryotes. RNA pol II contains a carboxy terminal domain 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. In addition, RNA pol II, in combination with several other polymerase subunits, form the DNA binding domain of the polymerase, a groove in which the DNA template is transcribed into RNA.

Immunogen: This RNA Pol II antibody was raised against a synthetic peptide YSPTSpPS corresponding to human RNA pol II.

**Buffer:** Purified IgG in 140 mM Hepes, pH 7.5, 70 mM NaCl, 32 mM NaOAc, 0.035% sodium azide, 30% glycerol. Sodium azide is highly toxic.

## **Application Notes:**

Applications Validated by Active Motif: ChIP-Seq: 4 µg per ChIP WB\*: 0.1 – 1 µg/ml IF: 2 µg/ml.

\*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.

AbFlex<sup>®</sup> recombinant antibodies are genetically derived from DNA sequences of parental hybridoma clones. For details on the parental clone, see Catalog No. 39097.

**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.

Application Key: ChIP = Chromatin Immunoprecipitation; FACS = Flow Cytometry; IF = Immunofluorescence; IHC = Immunohistochemistry; IP = Immunoprecipitation; WB = Western Blot





ChIP-Seq of AbFlex<sup>®</sup> RNA Pol II antibody (rAb).

ChIP was performed using AbFlex RNA Pol II recombinant antibody with 20  $\mu$ g chromatin from the HT1080 cell line and 4  $\mu$ g of antibody. ChIP DNA was sequenced on the Illumina HiSeq and 12 million sequence tags were mapped to identify RNA Pol II binding sites. The image shows binding across a region of chromosome 12.

	Western Blot of AbFlex <sup>®</sup> RNA Pol II antibody (rAb). Western Blot: 30 µg HeLa cell nuclear extract* was run on an SDS-PAGE gel and probed with AbFlex RNA Pol II (rAb) at 1 µg/mL. *Nuclear extract prepared with high salt / sonication protocol. Contact Technical Service for additional info. MW: 217 kDa
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Immunofluorescence stain of AbFlex<sup>®</sup> RNA Pol II antibody (rAb). RNA Pol II was detected in human HeLa cells in a 2-step staining protocol using AbFlex RNA Pol II (rAb) at 2  $\mu$ g/mL, followed by Anti-mouse IgG2a Dylight® 488.