

Pol II monoclonal antibody - Classic

Purified mouse monoclonal Antibody Catalog # ADN10297

Specification

Pol II monoclonal antibody - Classic - Product Information

Application	CHIP, WB
Primary Accession	<u>P24928</u>
Reactivity	Human, Mouse,
	Xenopus
Host	Mouse
Clonality	Monoclonal
Calculated MW	217176

Pol II monoclonal antibody - Classic - Additional Information

Gene ID 5430

Other Names

DNA-directed RNA polymerase II subunit RPB1, RNA polymerase II subunit B1, 2.7.7.6, DNA-directed RNA polymerase II subunit A, DNA-directed RNA polymerase III largest subunit, RNA-directed RNA polymerase II subunit RPB1, 2.7.7.48, POLR2A, POLR2

Target/Specificity Pol II

Precautions

Pol II monoclonal antibody - Classic is for research use only and not for use in diagnostic or therapeutic procedures.

Pol II monoclonal antibody - Classic - Protein Information

Name POLR2A (HGNC:9187)

Function

DNA-dependent RNA polymerase catalyzes the transcription of DNA into RNA using the four ribonucleoside triphosphates as substrates. Largest and catalytic component of RNA polymerase II which synthesizes mRNA precursors and many functional non-coding RNAs. Forms the



ChIP assays were performed using human HeLa cells, the Diagenode monoclonal antibody against Pol II (cat. No. ADN10297) and optimized PCR primer pairs for qPCR. ChIP was performed with the "iDeal ChIP-seq" kit (cat. No. C01010051), using sheared chromatin from 1 million cells. A titration consisting of 1, 2, 5 and 10 µl of antibody per ChIP experiment was analyzed. IgG (2 µg/ IP) was used as a negative IP control. Quantitative PCR was performed with primers specific for the promoter of the GAPDH and EIF4A2 genes, used as positive controls, and for the MYOD1 gene and the Sat2 satellite repeat, used as negative controls. Figure 1 shows the recovery, expressed as a % of input (the relative amount of immunoprecipitated DNA compared to input DNA after qPCR analysis).



polymerase active center together with the second largest subunit. Pol II is the central component of the basal RNA polymerase II transcription machinery. It is composed of mobile elements that move relative to each other. RPB1 is part of the core element with the central large cleft, the clamp element that moves to open and close the cleft and the jaws that are thought to grab the incoming DNA template. At the start of transcription, a single-stranded DNA template strand of the promoter is positioned within the central active site cleft of Pol II. A bridging helix emanates from RPB1 and crosses the cleft near the catalytic site and is thought to promote translocation of Pol II by acting as a ratchet that moves the RNA-DNA hybrid through the active site by switching from straight to bent conformations at each step of nucleotide addition. During transcription elongation, Pol II moves on the template as the transcript elongates. Elongation is influenced by the phosphorylation status of the C-terminal domain (CTD) of Pol II largest subunit (RPB1), which serves as a platform for assembly of factors that regulate transcription initiation, elongation, termination and mRNA processing. Regulation of gene expression levels depends on the balance between methylation and acetylation levels of tha CTD- lysines (By similarity). Initiation or early elongation steps of transcription of growth-factors-induced immediate early genes are regulated by the acetylation status of the CTD (PubMed:24207025). Methylation and dimethylation have a repressive effect on target genes expression (By similarity).

Cellular Location

Nucleus. Cytoplasm. Chromosome. Note=Hypophosphorylated form is mainly found in the cytoplasm, while the hyperphosphorylated and active form is nuclear (PubMed:26566685). Co-localizes with kinase SRPK2 and helicase DDX23 at chromatin loci where unscheduled R-loops form (PubMed:28076779).

Pol II monoclonal antibody - Classic -Protocols



Whole cell extracts (40 μ g) from HeLa cells transfected with Pol II siRNA (lane 2) and from an untransfected control (lane 1) were analysed by Western blot using the Diagenode antibody against Pol II (Cat. No. ADN10297) diluted 1:500 in TBS-Tween containing 5% skimmed milk. The position of the protein of interest is indicated on the right; the marker (in kDa) is shown on the left.



Provided below are standard protocols that you may find useful for product applications.

- Western Blot
- <u>Blocking Peptides</u>
- Dot Blot
- Immunohistochemistry
- Immunofluorescence
- Immunoprecipitation
- Flow Cytomety
- <u>Cell Culture</u>