# **Product Datasheet**

# RNA Polymerase II/POLR2A [p Ser5] Antibody (C.15200007) - BSA Free NBP2-59219

Unit Size: 50 ug

Store at -20C. Avoid freeze-thaw cycles.

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#### NBP2-59219

RNA Polymerase II/POLR2A [p Ser5] Antibody (C.15200007) - BSA Free

Product Information			
Unit Size	50 ug		
Concentration	Please see the vial label for concentration. If unlisted please contact technical services.		
Storage	Store at -20C. Avoid freeze-thaw cycles.		
Clonality	Monoclonal		
Clone	C.15200007		
Preservative	0.05% Sodium Azide		
Isotype	IgG1		
Purity	Protein A purified		
Buffer	PBS		
Product Description			
Host	Mouse		
Gene ID	5430		
Gene Symbol	POLR2A		
Species	Human, Zebrafish		
Reactivity Notes	Use in Zebrafish reported in secitific publication PMID: 32652076		
Immunogen	The exact immunogen is propietary information.		
Product Application Details			
Applications	Western Blot, ELISA, Immunocytochemistry/ Immunofluorescence, Chromatin Immunoprecipitation (ChIP), Chromatin Immunoprecipitation Sequencing, Knockdown Validated		
Recommended Dilutions	Western Blot 1:1000, ELISA 1:3000, Immunocytochemistry/ Immunofluorescence 1:500, Chromatin Immunoprecipitation (ChIP) 1-2 ug/IP, Chromatin Immunoprecipitation Sequencing 1 - 2 ug/ChIP, Knockdown Validated		

#### Images

Western Blot: RNA Polymerase II/POLR2A [p Ser5] Antibody (C.15200007) [NBP2-59219] - Whole cell extracts (40 ug) from HeLa cells transfected with Pol II siRNA (lane 2) and from an untransfected control (lane 1) were analysed by Western blot using the antibody against Pol II S5p diluted 1:1,000 in TBS-Tween containing 5% skimmed milk. The position of the protein of interest is indicated on the right; the marker (kDa) is shown on the left.

	1	2		
250	E.	3	- Pol II S5p	
130 —	5	2.		
95—	20			
72—				
55—				
36 <u></u> 28 <u></u>				

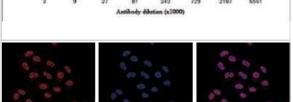


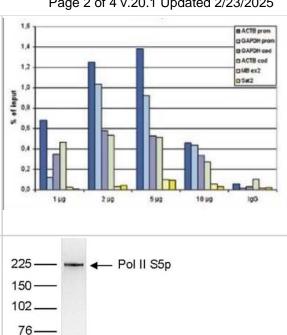
Chromatin Immunoprecipitation: RNA Polymerase II/POLR2A [p Ser5] Antibody (C.15200007) [NBP2-59219] - ChIP assays were performed using human HeLa cells, the antibody against Pol II S5p and optimized PCR primer pairs for qPCR. ChIP was performed with a ChIP-seq kit, using sheared chromatin from 1 million cells. A titration consisting of 1, 2, 5 and 10 ug of antibody per ChIP experiment was analyzed. IgG (2 ug/IP) was used as a negative IP control. Quantitative PCR was performed with primers specific for the promoter and the coding region of the constitutively expressed GAPDH and ACTB genes, used as positive controls, and for exon 2 of the inactive myoglobin (MB) gene and the Sat2 satellite repeat, used as negative controls. Figure shows the recovery, expressed as a % of input (the relative amount of immunoprecipitated DNA compared to input DNA after qPCR analysis).

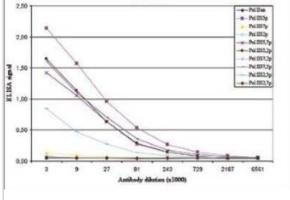
Western Blot: RNA Polymerase II/POLR2A [p Ser5] Antibody (C.15200007) [NBP2-59219] - Nuclear extracts (25 ug) from HeLa cells were analysed by Western blot using the antibody against Pol II S5p diluted 1:1,000 in TBS-Tween containing 5% skimmed milk. The position of the protein of interest is indicated on the right; the marker (kDa) is shown on the left.

ELISA: RNA Polymerase II/POLR2A [p Ser5] Antibody (C.15200007) [NBP2-59219] - Cross reactivity of the antibody directed against Pol IIS5p. To test the specificity an ELISA was performed using a serial dilution of the antibody against Pol IIS5p. The wells were coated with peptides containing the unmodified C-terminal repeat sequence as well as different phosphorylated peptides. Figure shows the specificity of the antibody for the S5 phosphorylation.

Immunofluorescence: RNA Polymerase II/POLR2A [p Ser5] Antibody (C.15200007) [NBP2-59219] - HeLa cells were stained with the antibody (left) diluted 1:500 in blocking solution followed by an anti-mouse antibody conjugated to Alexa Fluor 594. The middle panel shows staining of the nuclei with DAPI. A merge of the two stainings is shown on the right.







52.

38. 31



#### **Publications**

Lyu Y, Yang Y, Talwar V et al Hypoxia-inducible factor 1 recruits FACT and RNF20/40 to mediate histone ubiquitination and transcriptional activation of target genes Cell Rep 2024-03-22 [PMID: 38517892] (ChIP, Human)

Yang Y, Lu H, Chen C et al. HIF-1 Interacts with TRIM28 and DNA-PK to release paused RNA polymerase II and activate target gene transcription in response to hypoxia Nature communications 2022-01-14 [PMID: 35031618] (Chemotaxis)

Johansson JA, Marie KL, Lu Y et al. PRL3-DDX21 Transcriptional Control of Endolysosomal Genes Restricts Melanocyte Stem Cell Differentiation Dev. Cell 2020-07-08 [PMID: 32652076] (Chemotaxis, Zebrafish)

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# Products Related to NBP2-59219

HAF007	Goat anti-Mouse IgG Secondary Antibody [HRP]
NB720-B	Rabbit anti-Mouse IgG (H+L) Secondary Antibody [Biotin]
NBP1-97005-0.5mg	Mouse IgG1 Isotype Control (MG1)
H00005430-Q01-10ug	Recombinant Human RNA Polymerase II/POLR2A GST (N-Term) Protein

#### Limitations

This product is for research use only and is not approved for use in humans or in clinical diagnosis. Primary Antibodies are guaranteed for 1 year from date of receipt.

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