diagenode
A Hologic Company

TECHNICAL DATASHEET

p300 antibody

Cat. No. C15200211

Type: Monoclonal, **ChIP grade/ChIP-seq grade**
Source: Mouse
Lot: 002
Size: 10 µg / 50 µg
Concentration: 1.8 µg/µl

Specificity: Human: positive
Other species: not tested
Purity: Protein A purified monoclonal antibody
Storage: Store at -20°C; for long storage, store at -80°C. Avoid multiple freeze-thaw cycles.
Storage buffer: PBS containing 0.05% azide

Precautions: This product is for research use only. Not for use in diagnostic or therapeutic procedures.

Description: Monoclonal antibody raised in mouse against human p300 (E1A Binding Protein P300) by DNA immunization in which the C-terminal part of the protein was cloned and expressed.

Applications

Applications	Suggested dilution	References
ChIP/ChIP-seq*	2 - 5 µg per ChIP	Fig 1, 2
Western Blotting	not recommended	

*Please note that the optimal antibody amount per IP should be determined by the end-user. We recommend testing 1-10 µg per IP.

Target description

p300 (UniProt/Swiss-Prot entry Q09472) is a histone acetyltransferase that regulates transcription via chromatin remodelling. As such it is important for cell proliferation and differentiation. p300 is able to acetylate all four core histones in nucleosomes. Acetylation of histones is associated with transcriptional activation. p300 also acetylates non-histone proteins such as HDAC1 leading to its inactivation and modulation of transcription. It has also been identified as a co-activator of HIF1A (hypoxia-inducible factor 1 alpha), and thus plays a role in the stimulation of hypoxia-induced genes such as VEGF. Defects in the p300 gene are a cause of Rubinstein-Taybi syndrome and may also play a role in epithelial cancer.

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Results

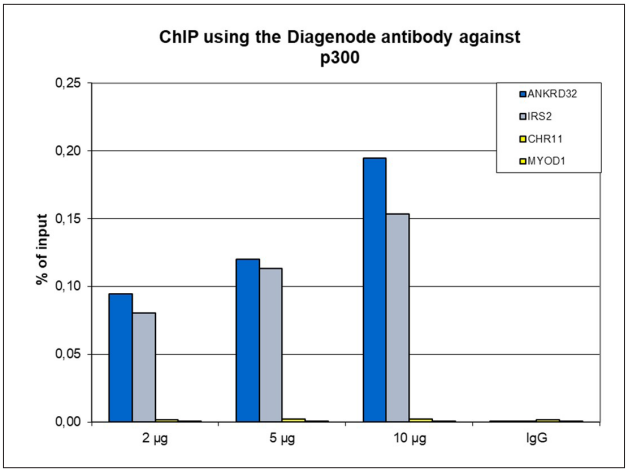

Figure 1. ChIP results obtained with the Diagenode monoclonal antibody directed against p300

ChIP was performed using HeLa cells, the Diagenode monoclonal antibody against p300 (cat. No. C15200211) and optimized PCR primer sets for qPCR. ChIP was performed with the "iDeal ChIP-seq" kit (cat. No. C01010055), using sheared chromatin from 4 million cells. A titration of the antibody consisting of 2, 5 and 10 µg per ChIP experiment was analysed. IgG (2 µg/IP) was used as negative IP control. Quantitative PCR was performed with primers for two genomic regions near the ANKRD32 and IRS2 genes, used as positive controls, and for the coding region of the inactive MYOD1 gene and an intergeic region on chromosome 11, 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).

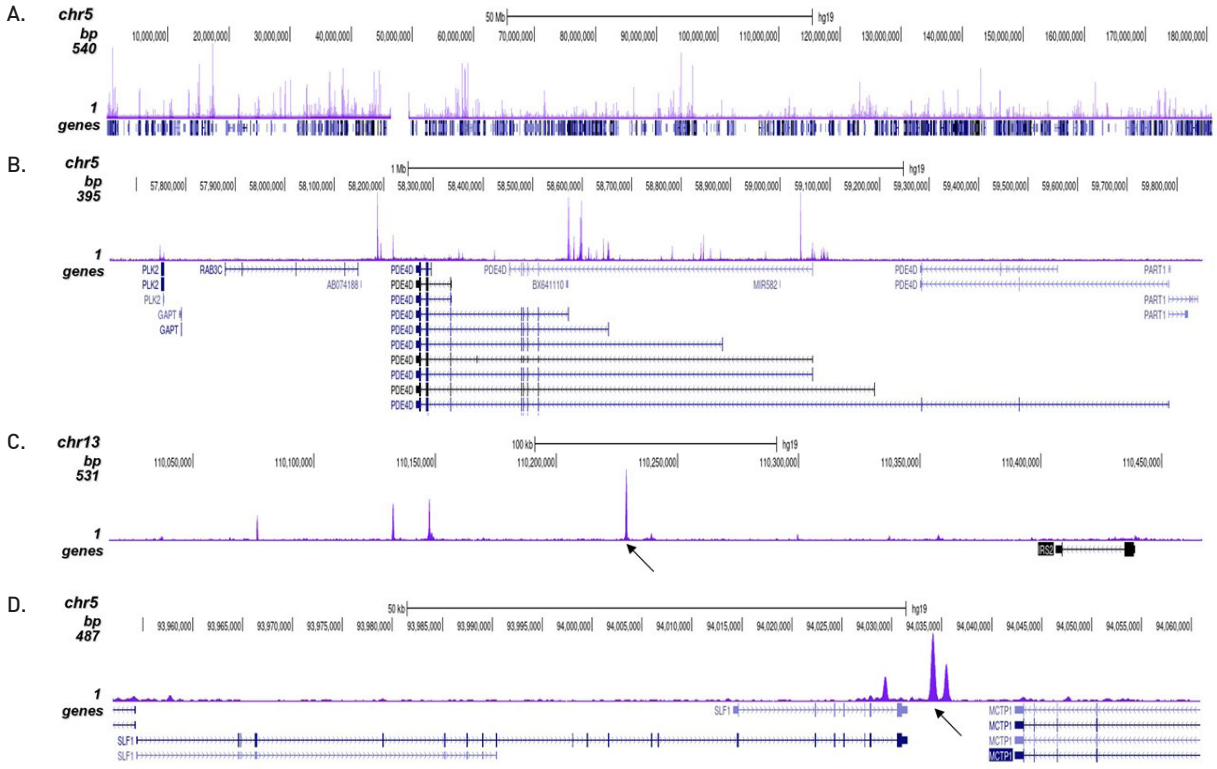

Figure 2. ChIP-seq results obtained with the Diagenode monoclonal antibody directed against p300

ChIP was performed with 5 µg of the Diagenode antibody against p300 (cat. No. C15200211) on sheared chromatin from 4 million HeLa cells as described above. The IP'd DNA was subsequently analysed on an Illumina NovaSeq. Library preparation, cluster generation and sequencing were performed according to the manufacturer's instructions. The 50 bp tags were aligned to the human genome using the BWA algorithm. Figure 2 shows the peak distribution along the complete sequence and a 3 mb region of chromosome 5 (figure 2A and B) and in two regions surrounding the IRS2 and ANKRD32 (SLF1) positive control genes (figure 2C and D). The position of the amplicon used for ChIP-qPCR is indicated by an arrow.