diagenode
A Hologic Company

TECHNICAL DATASHEET

# CBP polyclonal antibody - Classic

**Other name: CREBBP, KAT3A, RSTS**

**Cat. No.** C15410224
**Type:** Polyclonal **ChIP-grade/ChIP-seq grade**
**Source:** Rabbit
**Lot #:** 42844
**Size:** 25 µl/100 µl
**Concentration:** 0.66 µg/µl

**Specificity:** Human, mouse: positive
Other species: not tested
**Purity:** Affinity purified polyclonal antibody in 0.1 M Tris (pH 7) containing 0.1 M glycine, 10% glycerol and 0.01% thimerosal.
**Storage:** Store at -20°C; for long storage, store at -80°C. Avoid multiple freeze-thaw cycles.
**Precautions:** This product is for research use only. Not for use in diagnostic or therapeutic procedures.

**Description:** Polyclonal antibody raised in rabbit against CBP (CREB Binding Protein), using a recombinant protein.

## Applications

| | Suggested dilution/amount | References |
|---|---|---|
| ChIP* | 5 µg/ChIP | Fig 1, 2 |
| Western blotting | 1:500 - 1:3,000 | Fig 3 |
| Immunoprecipitation | 2.5 µg per IP | Fig 4 |
| Immunofluorescence | 1:100 - 1:1,000 | Fig 5 |
| Immunohistochemistry | 1:100 - 1:1,000 | Fig 6 |

*Please note that the optimal antibody amount per IP should be determined by the end-user. We recommend testing 1-5 µg per IP.

## Target description

CBP (UniProt/Swiss-Prot entry Q92793) acts as a histone acetylase thereby activating transcription. It also acts as a scaffold to stabilize additional protein interactions with the transcription complex and is involved in the transcriptional coactivation of many different transcription factors..CBP is also able to acetylate non-histone proteins such as NCOA3 and FOXO1. Further, CBP binds specifically to the cAMP-response element binding protein (CREB) and enhances its transcriptional activity toward cAMP-responsive genes. The latter play a critical role in embryonic development, growth control, and homeostasis by coupling chromatin remodelling to transcription factor recognition. Mutations in the CBP gene cause Rubinstein-Taybi syndrome (RTS) and chromosomal translocations involving this gene have been associated with acute myeloid leukemia.

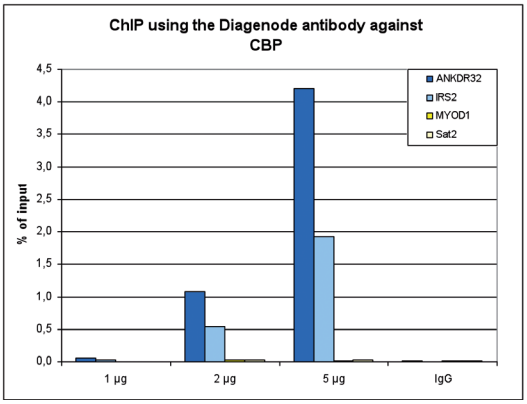


**Figure 1. ChIP results obtained with the Diagenode antibody directed against CBP**

ChIP assays were performed using HeLa cells, the Diagenode antibody against CBP (Cat. No. C15410224) and optimized 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 1, 2 and 5 µg per ChIP experiment was analysed. IgG (1 µg/IP) was used as negative IP control. QPCR was performed with primers for the ANKRD32 and IRS2 genes, used as positive controls, and for the MYOD1 gene and the Sat2 satellite repeat, used as a 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).

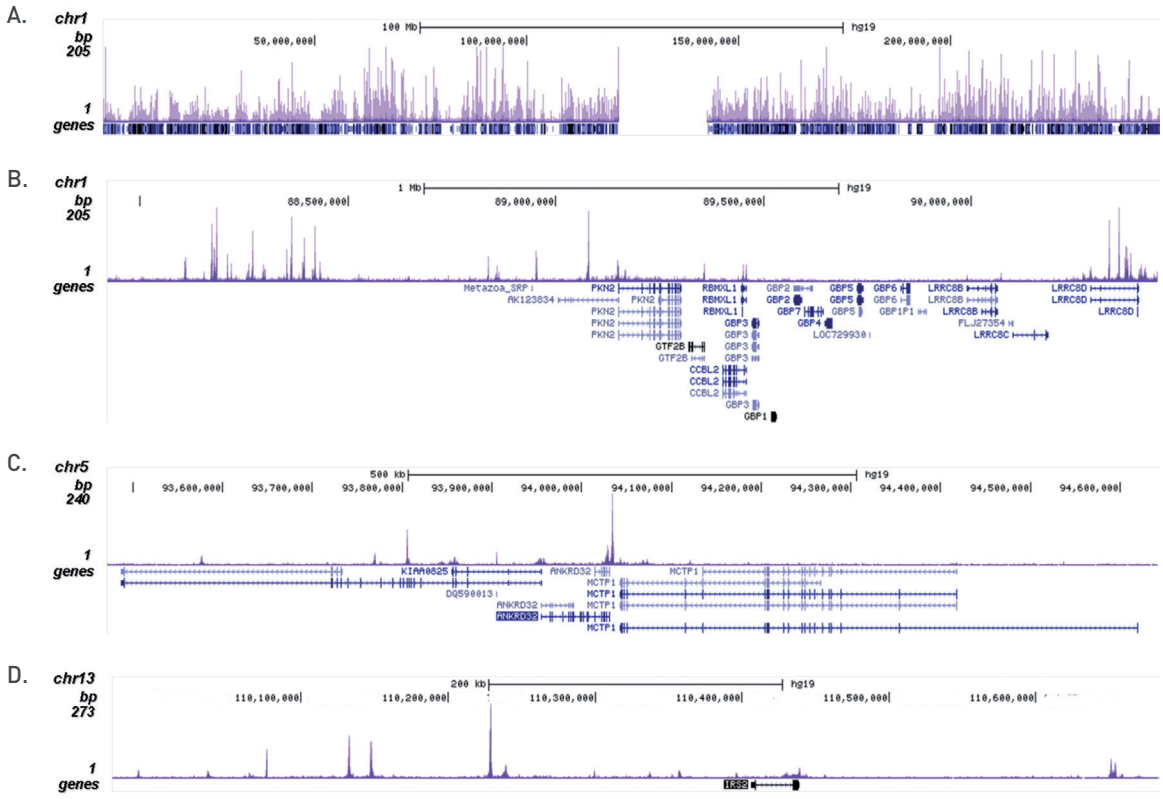


**Figure 2. ChIP-seq results obtained with the Diagenode antibody directed against CBP**

ChIP was performed on sheared chromatin from 4 million HeLa cells using 5 µg of the Diagenode antibody against CBP (Cat. No. C15410224) as described above. The IP'd DNA was subsequently analysed on an Illumina HiSeq. 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 enrichment along the complete sequence and an 2 Mb region of human chromosome 1 (fig 2A and B), and in two genomic regions surrounding the ANKRD32 and IRS2 positive control genes (fig 2C and D).


**Figure 3. Western blot analysis using the Diagenode antibody directed against CBP**

Whole cell extracts from H1299 cells (30 µg) were analysed by Western blot using the Diagenode antibody against CBP (Cat. No. C15410224) diluted 1:1,000. The position of the protein of interest is indicated on the right; the marker (in kDa) is shown on the left.

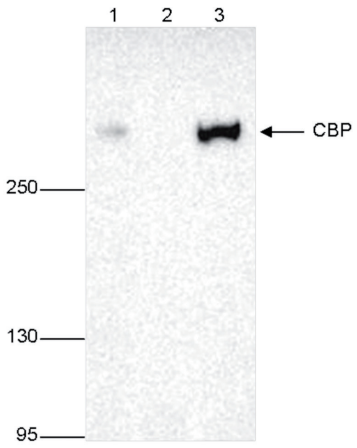


**Figure 4. Immunoprecipitation using the Diagenode antibody directed against CBP**

Immunoprecipitation was performed on whole cell extracts from HeLa cells (1 mg) using 2.5 µg of the Diagenode antibody against CBP (Cat. No. C15410224). An equal amount of rabbit IgG was used as a negative control. The immunoprecipitated CBP protein was detected by western blot with the CBP antibody diluted 1:1,000. The IP with the CBP antibody and with the IgG negative control are shown in lane 3 and lane 2, respectively. Lane 1 shows the input (40 µg of HeLa whole cell extract).

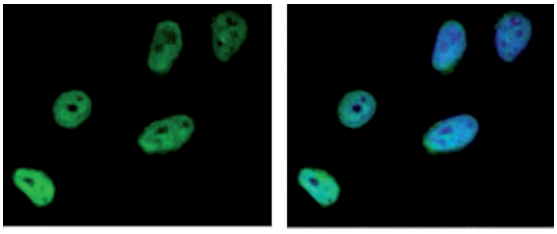

**Figure 5. Immunofluorescence with the Diagenode antibody directed against CBP**

HeLa cells were fixed with formaldehyde and stained with the Diagenode antibody against CBP (Cat. C15410224) diluted 1:200 (left). The right picture shows costaining with Hoechst 33342 nucleic acid stain.

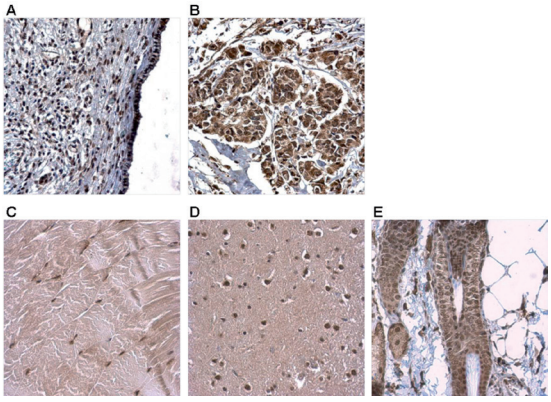


**Figure 6. Immunohistochemistry with the Diagenode antibody directed against CBP**

Formalin fixed paraffin embedded human cervical carcinoma (fig 6A) or breast carcinoma (fig 6B) tissue or mouse muscle (fig 6C), forebrain (fig 6D) and skin (fig 6E) tissue was stained with the Diagenode antibody against CBP (cat. No. C15410224) diluted 1:750 followed by a peroxidase labelled goat anti-rabbit secondary antibody.

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**Last update:** october 22, 2015