diagenode
Innovating Epigenetics Solutions

TECHNICAL DATASHEET

TBP monoclonal antibody - Classic

Cat. No. C15200002

Type: Monoclonal **Chip grade, ChIP-seq grade**	Specificity: Human, mouse
Isotype: NA	Concentration: 8 µg/µl
Source: Mouse	Purity: Ammonium sulphate purified
Lot No.: DA-0010	Storage:
Size: 100 µg	Precautions: This product is for research use only. Not for use in diagnostic or therapeutic procedures.

Last Data Sheet Update: January 17, 2017

Description

Alternative names: **GTF2D**, **GTF2D1**, **SCA17**, **TF2D**, **TFIID**

Monoclonal antibody raised in mouse against the amino-terminal domain of human TBP (TATA box binding protein).

Applications

Applications	Suggested dilution	References
ChIP *	4 - 5 µg/IP	Fig 1, 2
WB	1:500	Fig 3

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

Target Description

Monoclonal antibody raised in mouse against the amino-terminal domain of human TBP (TATA box binding protein).

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Validation Data

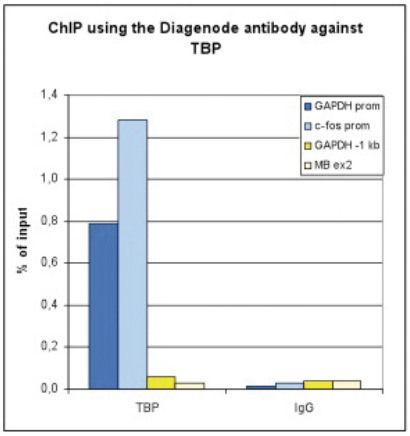
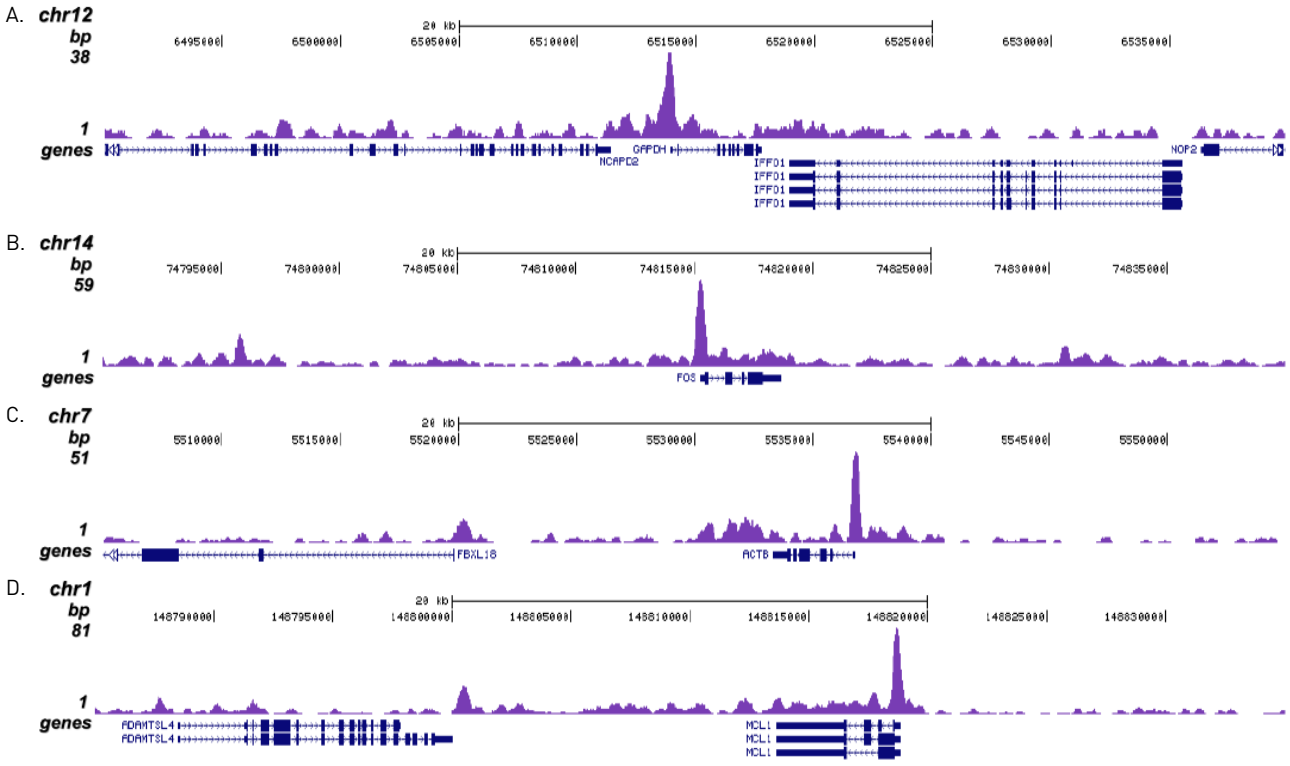


Figure 1 ChIP-seq results obtained with the Diagenode monoclonal antibody directed against TBP

ChIP was performed with 5 µg of the Diagenode antibody against TBP (Cat. No. C15200002) on sheared chromatin from 1 million HeLaS3 cells using the "Auto Histone ChIP-seq" kit (Cat. No. C01010022) on the IP-Star automated system. The IP'd DNA was analysed by QPCR with optimized PCR primer pairs for the promoters of the active GAPDH and c-fos genes, used as positive control targets, and for a region 1 kb upstream of the GAPDH promoter and the coding region of the inactive MB gene, used as negative control targets (figure 2A). The IP'd DNA was subsequently analysed with an Illumina Genome Analyzer. Library preparation, cluster generation and sequencing were performed according to the manufacturer's instructions. The 36 bp tags were aligned to the human genome using the ELAND algorithm. Figure 2 shows the peak distribution in 50 kb regions surrounding the GAPDH, c-fos, ACTB and MCL1 genes (figure 2B, C, D and E, respectively). These results clearly show a localisation of TBP at the promoters of actively transcribed genes.



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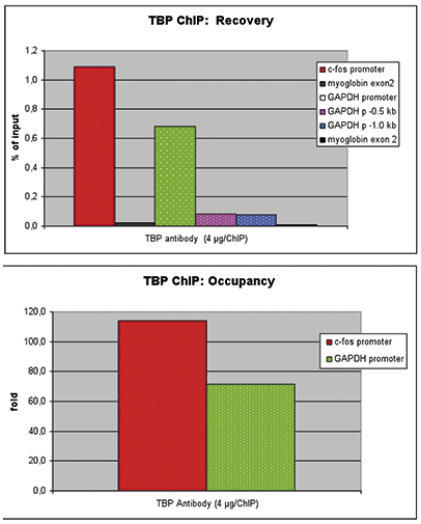

Figure 2 ChIP results obtained with the Diagenode monoclonal antibody against TBP

ChIP assays were performed using U2OS cells, the Diagenode antibody directed against TBP (Cat. No. C15200002) and optimized primer sets for qPCR. Sheared chromatin from 1x10e6 cells and 4 µg of antibody were used per ChIP experiment. QPCR was performed with primers for the promoter of the c-fos and GAPDH genes (Cat. No. C17011004 and C17011001), a region 0.5 and 1 kb upstream of the GAPDH promoter (Cat. No. C17011002 and C17011003), respectively, and for exon 2 of the myoglobin gene (cat. No. C17011006) as a negative control. Figure 1 shows the recovery (the relative amount of immunoprecipitated DNA compared to input DNA) and the occupancy (ratio +/- control target). These results demonstrate the occupancy of both promoters by TBP.

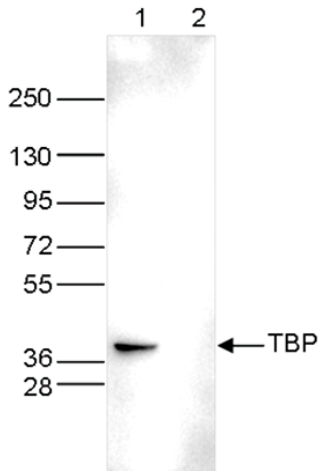

Figure 3. Western blot analysis using the Diagenode monoclonal antibody directed against TBP

Whole cell extracts (40 µg) from HeLa cells transfected with TBP siRNA (lane 2) and from an untransfected control (lane 1) were analysed by Western blot using the Diagenode antibody against TBP (Cat. No. C15200002) diluted 1:500 in TBSTween 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.

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