diagenode
A Hologic Company

TECHNICAL DATASHEET

FOXA2 polyclonal antibody

Other names: HNF3B, TFC3B, HNF-3B, TCF-3B

Cat. No. C15410343
Type: Polyclonal **ChIP grade/ChIP-seq grade**
Source: Rabbit
Lot #: A2683-0040
Size: 50 µg/21 µl
Concentration: 2.4 µg/µl

Specificity: Human: positive
Other species: not tested
Purity: Affinity purified polyclonal antibody in PBS containing 0.05% azide.
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 human FOXA2 (Forkhead Box A2), using two synthetic peptides containing a sequence from the central part and the N-terminus of the protein, respectively.

Applications

Applications	Suggested dilution	References
ChIP*	2 µg per ChIP	Fig 1, 2
ELISA	1:5,000	Fig 3
WB	1:500	Fig 4

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

Target description

FOXA2 (UniProtKB/Swiss-Prot entry Q9Y261) is a member of the forkhead class of transcription factors which are involved in the transcriptional activation of liver-specific genes such as AFP, albumin, tyrosine aminotransferase, PEPCK and transthyretin. FOXA2 also plays a role glucose homeostasis and in the regulation of fat metabolism and is required for the embryonic development of multiple endoderm-derived organ systems such as the liver, pancreas and lungs. The FOXA2 gene has been linked to sporadic cases of maturity-onset diabetes of the young.

Results

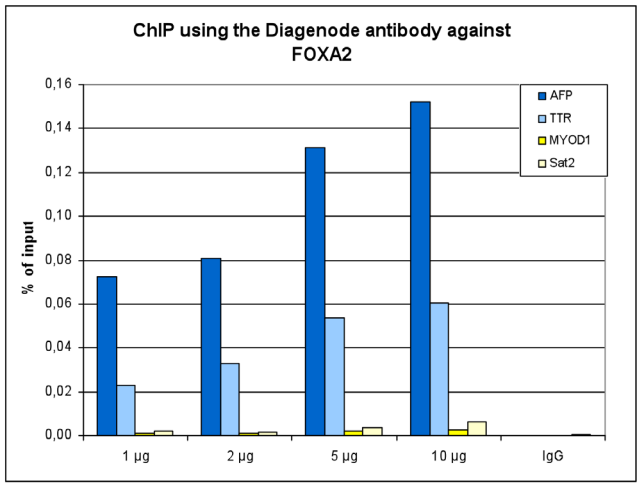

Figure 1. ChIP results obtained with the Diagenode antibody directed against FOXA2

ChIP assays were performed using HepG2 cells, the Diagenode antibody against FOXA2 (cat. No. C15410343) 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 consisting of 1, 2, 5 and 10 µg of antibody per ChIP experiment was analyzed. IgG (2 µg/IP) was used as a negative IP control. Quantitative PCR was performed with primers for the promoters of the AFP and TTR 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).

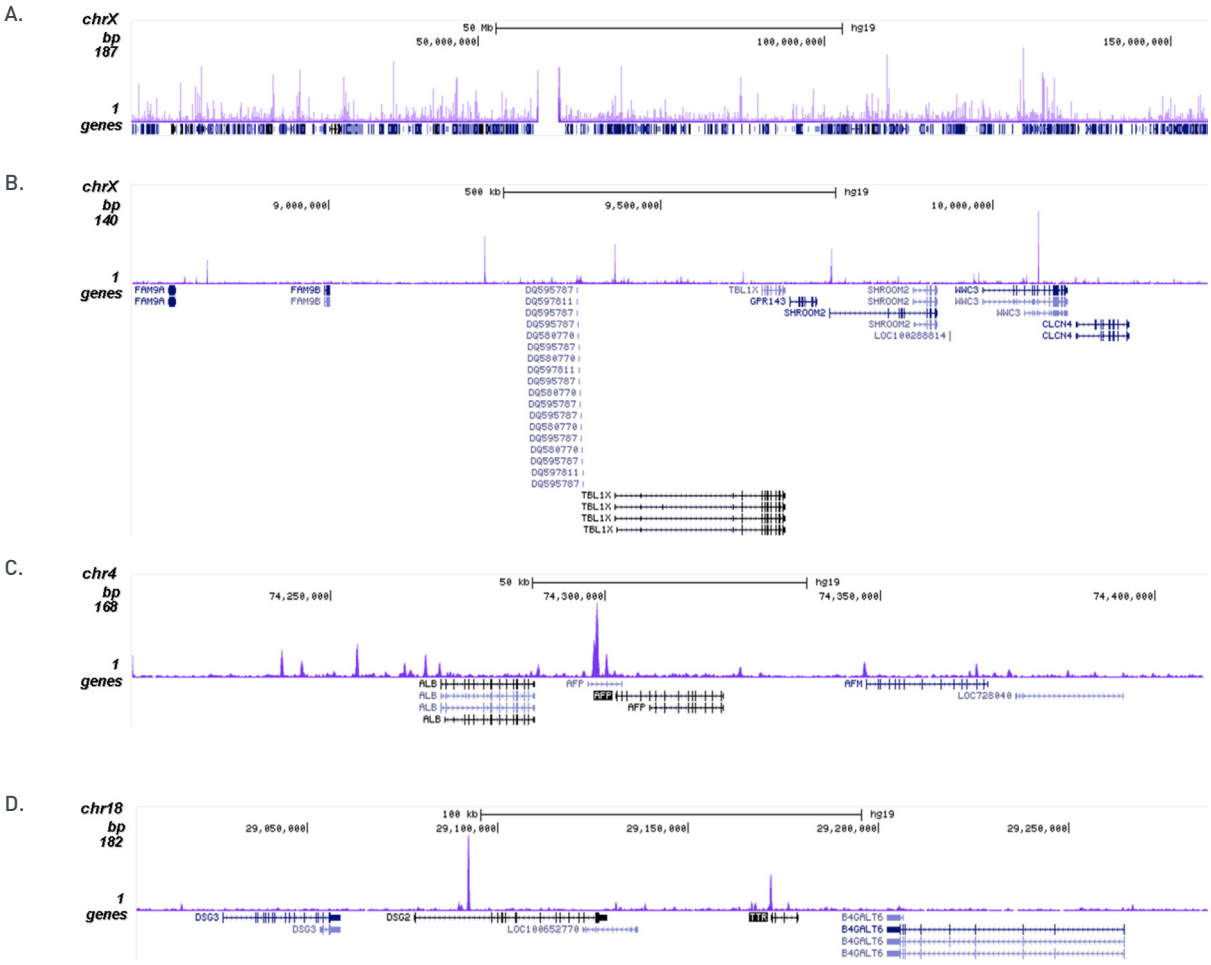

Figure 2. ChIP-seq results obtained with the Diagenode antibody directed against FOXA2

ChIP was performed with 2 µg of the Diagenode antibody against FOXA2 (Cat. No. C15410343) on sheared chromatin from 4,000,000 HepG2 cells using the "iDeal ChIP-seq" kit as described above. The IP'd DNA was subsequently analysed on an Illumina HiSeq 4000. 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 signal distribution along the complete sequence and a 1.5 Mb region of the human X-chromosome (figures 2A and B), and in two genomic regions surrounding the AFP and TTR positive control genes (figure 2C and D).

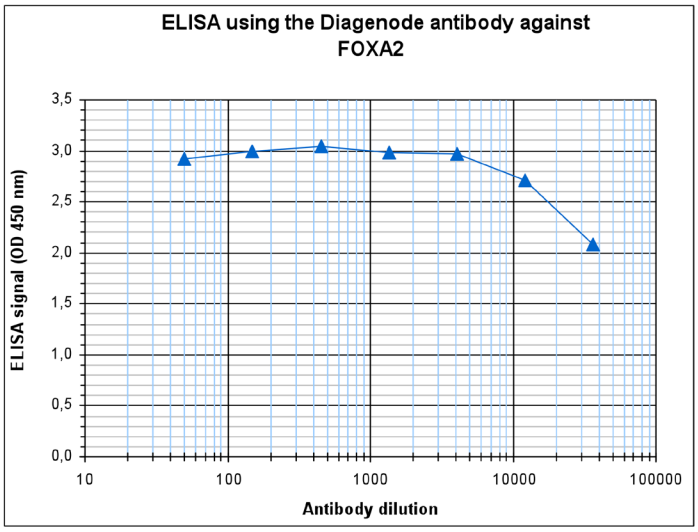

Figure 3. Determination of the antibody titer

To determine the titer of the antibody, an ELISA was performed using a serial dilution of Diagenode antibody directed against FOXA2 (Cat. No. C15410343). The plates were coated with the peptides used for immunization of the rabbit. By plotting the absorbance against the antibody dilution (Figure 3), the titer of the antibody was estimated to be 1:176,000.

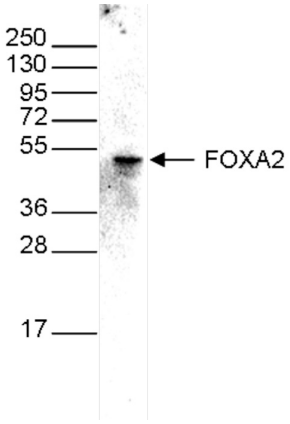

Figure 4. Western blot analysis using the Diagenode antibody directed against FOXA2

Whole cell extracts from HepG2 cells were analysed by Western blot using the Diagenode antibody against FOXA2 (Cat. No. C15410343) 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.

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