



TECHNICAL DATASHEET

ZHX2 polyclonal antibody - Classic

Other name: AFR1, RAF

Cat. No. C15410260

Type: Polyclonal ChIP-grade, ChIP-seq grade

Source: Rabbit **Lot #:** 40107 **Size:** 25 µl/100 µl

Concentration: 0.86 µg/µl

Specificity: Human: positive

Purity: Affinity purified polyclonal antibody in 0.1 M Tris-HCl 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 ZHX2 (Zinc Fingers And Homeoboxes 2), using a recombinant protein.

Applications

	Suggested dilution*	Results
ChIP*	2 - 5 μg per ChIP	Fig 1, 2
Western blotting	1:500 - 1:3,000	Fig 3
Immunofluorescence	1:100 - 1:1,000	Fig 4
Immunohistochemistry	1:100 - 1:1,000	Fig 5

^{*} 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

ZHX2 (UniProt/Swiss-Prot entry Q9Y6X8) is a transcriptional repressor that interacts with the A subunit of nuclear factor-Y (NF-YA). ZHX2 represses the promoter activity of the CDC25C gene.





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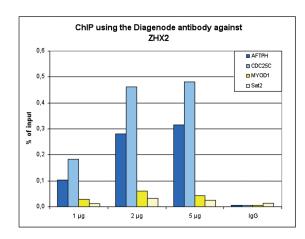
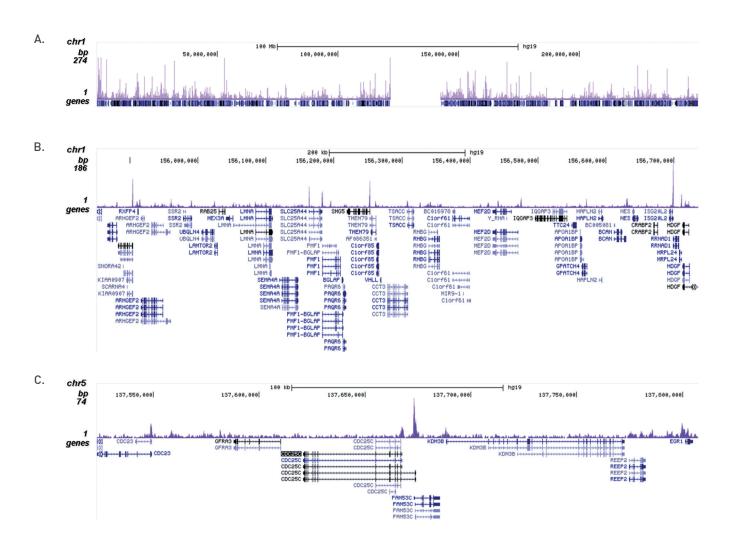


Figure 1. ChIP results obtained with the Diagenode antibody directed against ZHX2

ChIP assays were performed using HepG2 cells, the Diagenode antibody against ZHX2 (Cat. No. C15410260) 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 μ g per ChIP experiment was analysed. IgG (1 μ g/IP) was used as negative IP control. QPCR was performed with primers for the CDC25C and AFTPH promoters, 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).



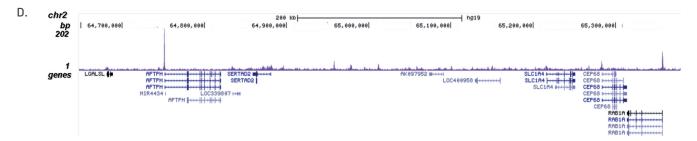


Figure 2. ChIP-seq results obtained with the Diagenode antibody directed against ZHX2

ChIP was performed on sheared chromatin from 5 million HepG2 cells using 5 µg of the Diagenode antibody against ZHX2 (Cat. No. C15410260). 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 a 1 Mb region of chromosomes 1 (fig 2A and B) and in two genomic regions surrounding the CDC25C and AFTPH genes on chromosome 5 and 2, respectively (fig 2C and D).

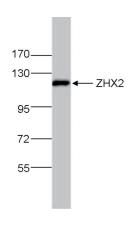


Figure 3. Western blot analysis using the Diagenode antibody directed against ZHX2

Whole cell extracts from HepG2 cells (30 µg) were analysed by Western blot using the Diagenode antibody against ZHX2 (Cat. No. C15410260) 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.

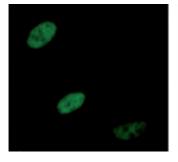




Figure 4. Immunofluorescence with the Diagenode antibody directed against ZHX2

HeLa cells were fixed with 4% formaldehyde for 15' at room temperature and stained with the Diagenode antibody against ZHX2 (Cat. C15410260) diluted 1:500 (left). The right picture shows costaining with Hoechst 33342 nucleic acid stain.

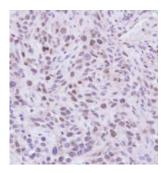


Figure 5. Immunohistochemistry using the Diagenode antibody directed against ZHX2

Formalin fixed paraffin embedded SAS xenograft tissue was stained with the Diagenode antibody against ZHX2 (Cat. No. C15410260) diluted 1:100 followed by a peroxidase labelled goat anti-rabbit secondary antibody.

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