

NFYA polyclonal antibody - Classic

Cat. No. **C15310261**

Type: Polyclonal ChIP grade, ChIP-seq grade	Specificity: Human
Isotype: NA	Concentration: not determined
Source: Rabbit	Purity: Whole antiserum
Lot No.: 001	Storage:
Size: 100 µl	Precautions: This product is for research use only. Not for use in diagnostic or therapeutic procedures.

Last Data Sheet Update: January 16, 2017

Description

Alternative names: **NF-YA, HAP2, CBF-A, CBF-B**

Polyclonal antibody raised in rabbit against NFYA (Nuclear transcription factor-Y, subunit A), using a KLH conjugated synthetic peptide from the N-terminus of the protein.

Applications

Applications	Suggested dilution	References
ChIP *	1:1,000	Fig 1, 2
Western Blotting	1:1,000	Fig 3
Immunohistochemistry	1:400	Fig 4

* 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

NFYA (UniProt/Swiss-Prot entry P23511) is one of the 3 subunits of the ubiquitous transcription factor NFY. All three subunits A, B and C are required to form a NFY-DNA complex. Depending on the cofactors involved, NFY can function both as an activator or a repressor, The NFYA subunit is responsible for the sequence specific interactions of the complex, suggesting a role as regulatory subunit. NFYA regulates the transcription of the core clock component ARNTL/BMAL1.

Validation data

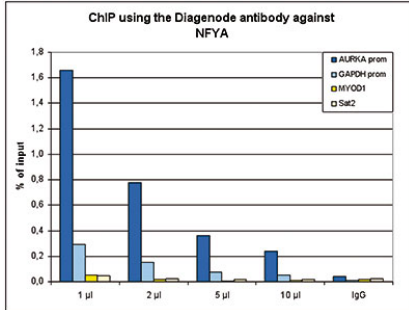


Figure 1. ChIP results obtained with the Diagenode antibody directed against NFYA

ChIP assays were performed using HeLa cells, the Diagenode antibody against NFYA [Cat. No. C15310261] 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, 5 and 10 µl per ChIP experiment was analysed. IgG (2 µg/IP) was used as negative IP control. QPCR was performed with primers for the AURKA and GAPDH 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).

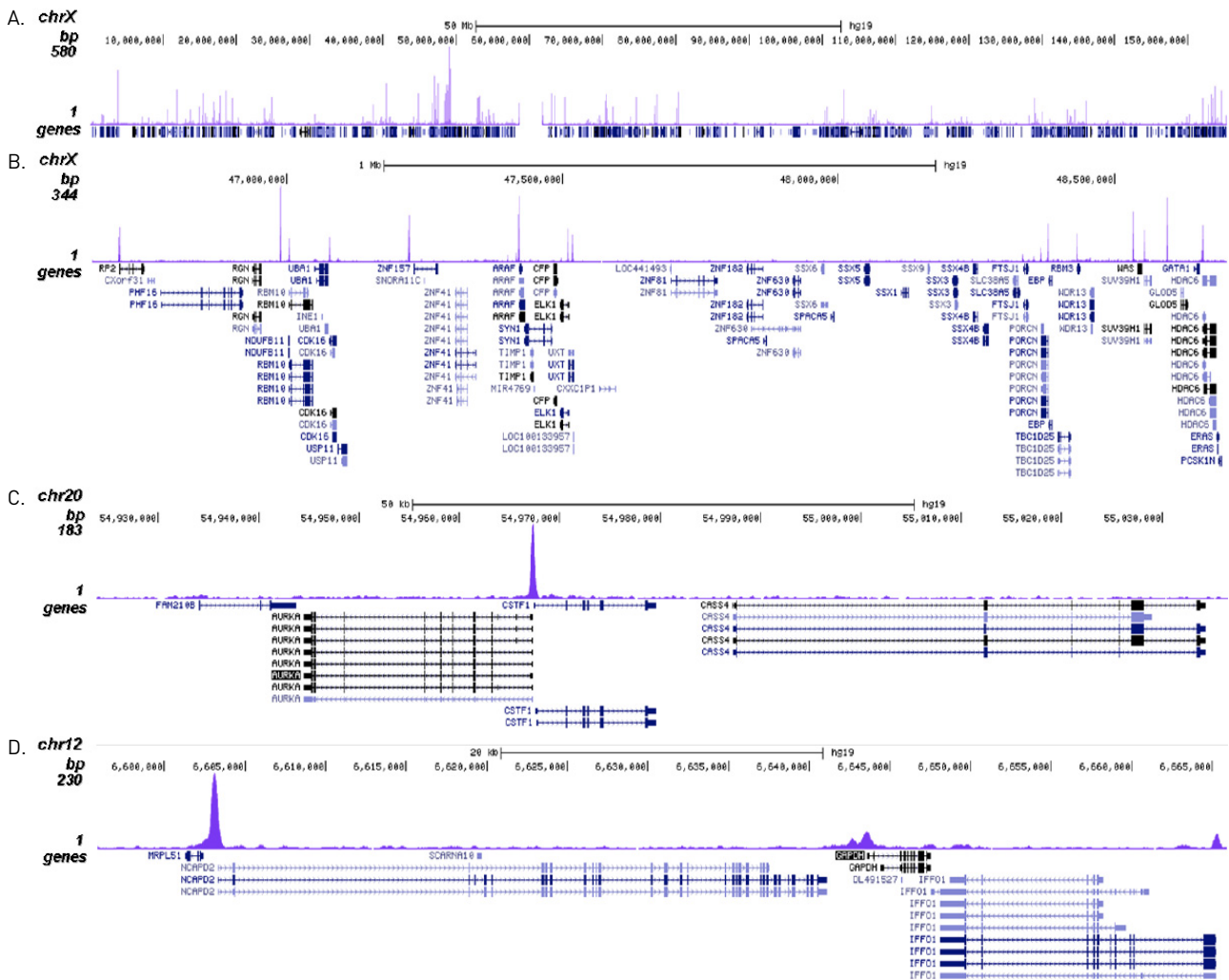


Figure 2. ChIP-seq results obtained with the Diagenode antibody directed against NFYA

ChIP was performed on sheared chromatin from 4 million HeLa cells using 1 µl of the Diagenode antibody against NFYA [Cat. No. C15310261] 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 a 2 Mb region of the human X chromosome (fig 2A and B), and in a two genomic regions surrounding the AURKA and GAPDH

positive control genes.

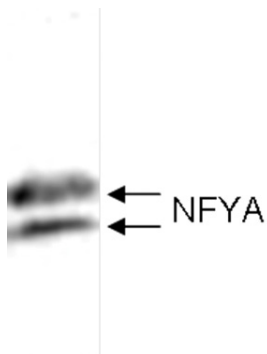


Figure 3. Western blot analysis using the Diagenode antibody directed against NFYA

Nuclear extracts from CHO-7 cells (35 µg) were analysed by Western blot using the Diagenode antibody against NFYA (Cat. No. C15310261) diluted 1:1,000. Figure 3 shows a doublet representing both isoforms of NFYA.

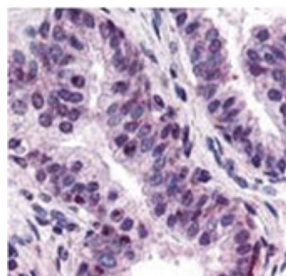


Figure 4. Immunohistochemistry using the Diagenode antibody directed against NFYA

Formalin fixed paraffin embedded human prostate tissue was stained with the Diagenode antibody against NFYA (cat. No. C15310261) diluted 1:400, followed by a peroxidase labelled anti-rabbit antibody. Figure 2 shows nuclear staining of NFYA (red).