diagenode
A Hologic Company

ChIP-seq grade

TECHNICAL DATASHEET

WDR5 polyclonal antibody

Other names: BIG3, SWD3

Cat. No. C15410027
Type: Polyclonal **ChIP grade / ChIP-seq grade**
Source: Rabbit
Lot #: 001
Size: 50 µg/25 µl
Concentration: 2.0 µg/µl

Specificity: Human: positive
Other species: not tested
Purity: Protein G purified polyclonal antibody in PBS containing 0.05% azide and 0.05% ProClin 300.
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 WDR5 (WD (tryptophan-aspartate) repeat domain 5), using a recombinant protein.

Applications

	Suggested dilution	Results
ChIP*	2 µg per ChIP	Fig 1, 2
Western blotting	1:1,000	Fig 3
Immunofluorescence	1:1,000	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

WDR5 (UniProtKB/Swiss-Prot entry P61964) belongs to the family of WD repeat proteins which are involved in different cellular processes such as cell cycle progression, signal transduction, apoptosis, and gene regulation. It is a component of the Set1A and Set1B histone H3 methylation complexes. These complexes methylate lysine 4 of H3, thereby activating gene transcription. WDR5 interacts with H3 dimethyl K4, but not with tri or mono methylated H3K4.

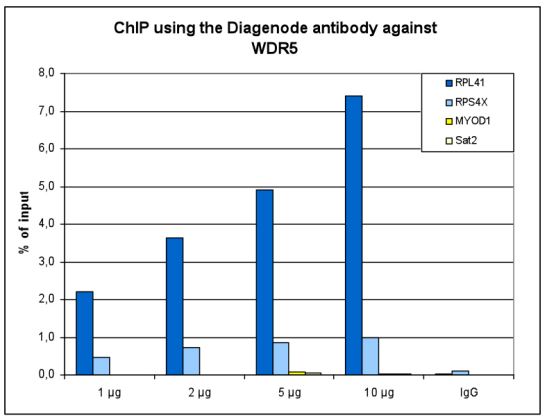

Figure 1. ChIP results obtained with the Diagenode antibody directed against WDR5

ChIP assays were performed using HeLa cells, the Diagenode antibody against WDR5 (Cat. No. C15410027) 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 optimized primers for the promoters of the RPL41 and RPS4X ribosomal protein 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).

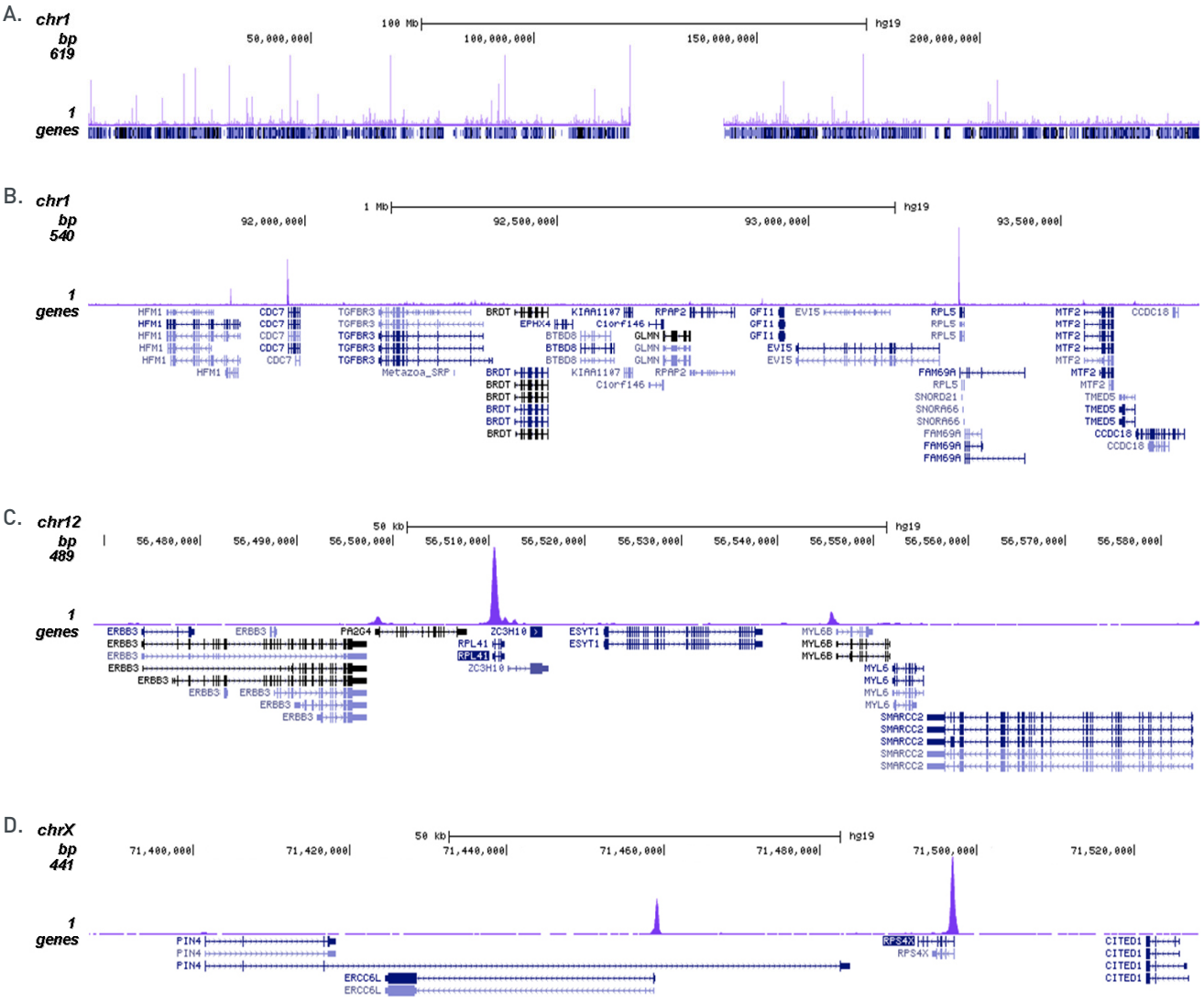

Figure 2. ChIP-seq results obtained with the Diagenode antibody directed against WDR5

ChIP was performed on sheared chromatin from 4 million HeLa cells using 2 µg of the Diagenode antibody against WDR5 (Cat. No. C15410027) 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 human chromosome 1 (fig 2A and B), and in two genomic regions surrounding the RPL41 and RPS4X positive control genes (fig 2C and D).

Results

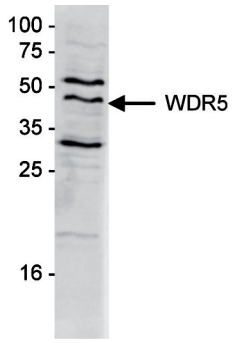

Figure 3. Western blot analysis using the Diagenode antibody directed against WDR5

Nclear extracts from HeLa cells (20 µg) were analysed by Western blot using the Diagenode antibody against WDR5 (Cat. No. C15410027) diluted 1:1,000 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.

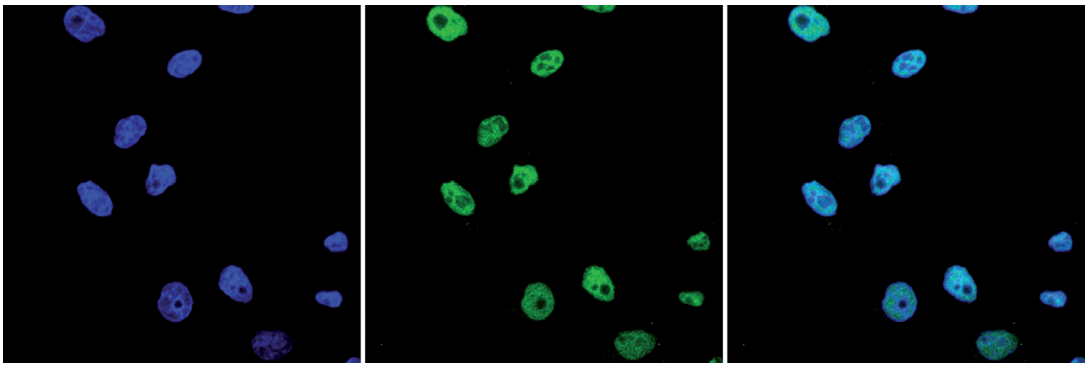

Figure 4. Immunofluorescence using the Diagenode antibody directed against WDR

HeLa cells were stained with the Diagenode antibody against WDR5 (Cat. No. C15410027) and with DAPI. Cells were fixed with 4% formaldehyde for 10' and blocked with PBS/TX-100 containing 5% normal goat serum and 1% BSA. The cells were immunofluorescently labeled with the WDR5 antibody (middle) diluted 1:1,000 in blocking solution followed by an anti-rabbit antibody conjugated to Alexa488. The left panel shows staining of the nuclei with DAPI. A merge of the two stainings is shown on the right.

Diagenode sa. BELGIUM | **EUROPE**
LIEGE SCIENCE PARK
Rue Bois Saint-Jean, 3
4102 Seraing (Ougrée) - Belgium
Tel: +32 4 364 20 50
Fax: +32 4 364 20 51
orders@diagenode.com
info@diagenode.com

Diagenode Inc. USA | **NORTH AMERICA**
400 Morris Avenue, Suite 101
Denville, NJ 07834 - USA
Tel: +1 862 209-4680
Fax: +1 862 209-4681
orders.na@diagenode.com
info.na@diagenode.com

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