diagenode
A Hologic Company

TECHNICAL DATASHEET

LSD1 polyclonal antibody

Other names: BHC110, AOF2, EC1, KDM1

Cat. No. C15410067
Type: Polyclonal **ChIP grade / ChIP-seq grade**
Source: Rabbit
Lot #: A57-00234P
Size: 50 µg/ 27 µl
Concentration: 2.2 µg/µl

Specificity: Human: positive. Other species: not tested.
Purity: Affinity 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 LSD1 (Lysine-specific demethylase 1), using a KLH-conjugated synthetic peptide from the inner part of the protein.

Applications

	Suggested dilution	References
ChIP *	1 µg/ChIP	Fig 1, 2
ELISA	1:1,000 - 1:10,000	Fig 3
Western blotting	1:4,000	Fig 4, 5
Immunofluorescence	1:200	Fig 6

* 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

LSD1 (lysine specific demethylase 1, UniProt/Swiss-Prot entry O60341) is a component of the histone demethylase complex that uses FAD as a prosthetic goup. LSD1 may have a dual effect on gene transcription. As it demethylates the mono- and dimethylated 'Lys-4' of histone H3, which are associated with transcriptional activation, LSD1 can act as a repressor of gene expression. However, LSD1 is also capable of demethylating 'Lys-9' of histone H3, a specific tag for epigenetic transcriptional repression, thereby leading to activation of androgen receptor target genes. LSD1 therefore mediates different processes such as embryonic development, cell differentiation and proliferation, stem and cancer cell biology.

Results

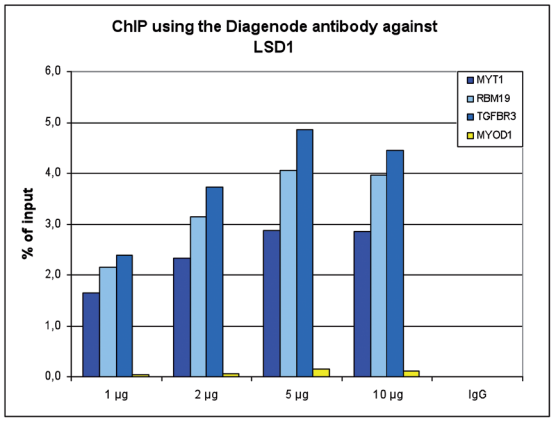

Figure 1. ChIP results obtained with the Diagenode antibody directed against LSD1

ChIP was performed with the Diagenode antibody against LSD1 (Cat. No. C15410067) on sheared chromatin from 4,000,000 K562 cells using the "iDeal ChIP-seq" kit (Cat. No. C01010055). An antibody titration consisting of 1, 2, 5 and 10 µg per ChIP experiment was analysed. IgG (2 µg/IP) was used as negative IP control. QPCR was performed with primers for specific regions in the MYT1, RBM19, and TGFBR3 genes, used as positive controls, and for the MYOD1 gene, used as negative control. Figure 1 shows the recovery, expressed as a % of input (the relative amount of immunoprecipitated DNA compared to input DNA after qPCR analysis).

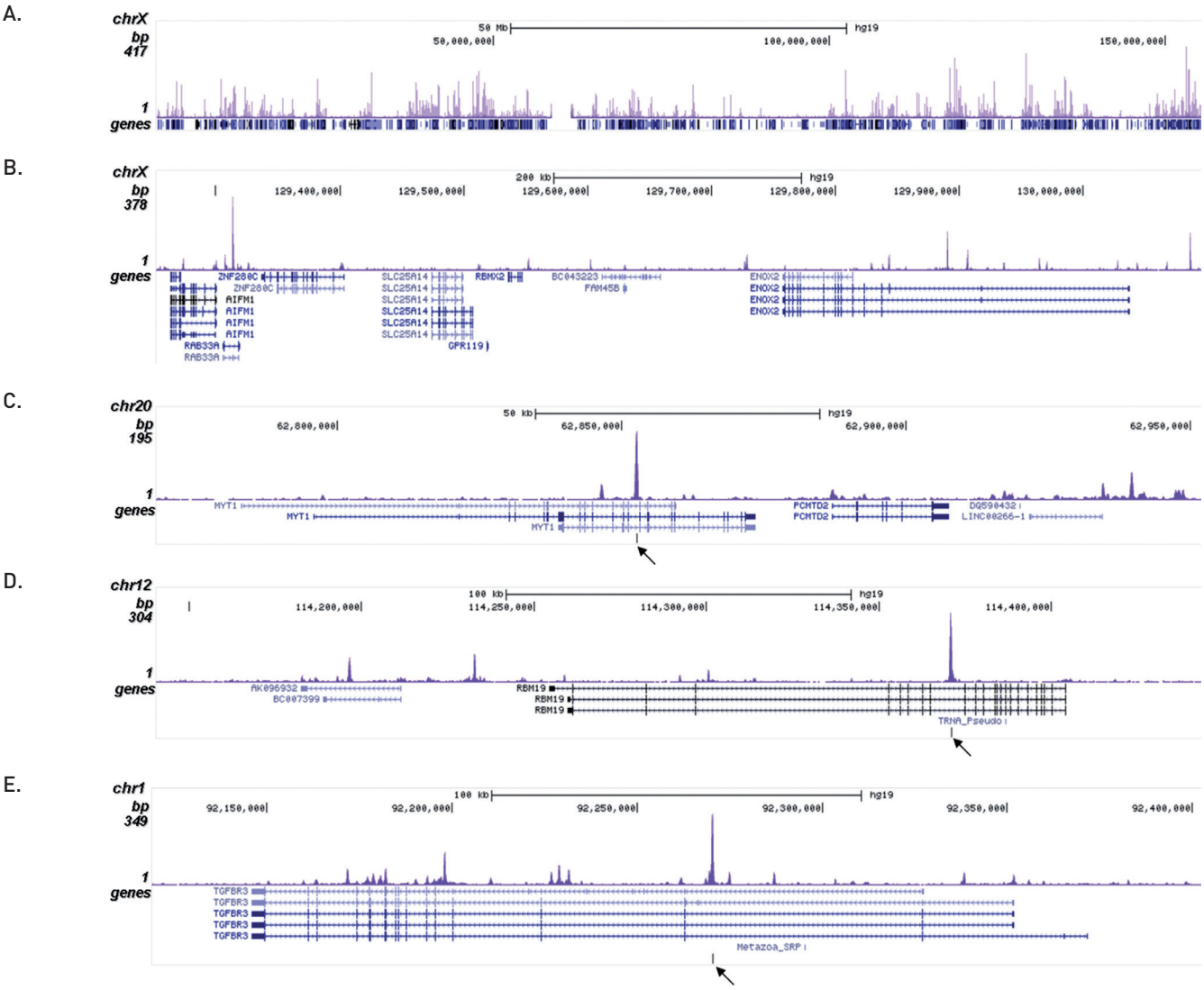

Figure 2. ChIP-seq results obtained with the Diagenode antibody directed against LSD1

ChIP was performed on sheared chromatin from 4,000,000 K562 cells using 1 µg of the Diagenode antibody against LSD1 (Cat. No. C15410067) 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 peak distribution along the complete sequence and a 600 kb region of the X-chromosome (figure 2A and B) and in three regions surrounding the MYT1, RBM19 and TGFBR3 positive control genes, respectively (figure 2C, D and E). The position of the amplicon used for ChIP-qPCR is indicated by an arrow.

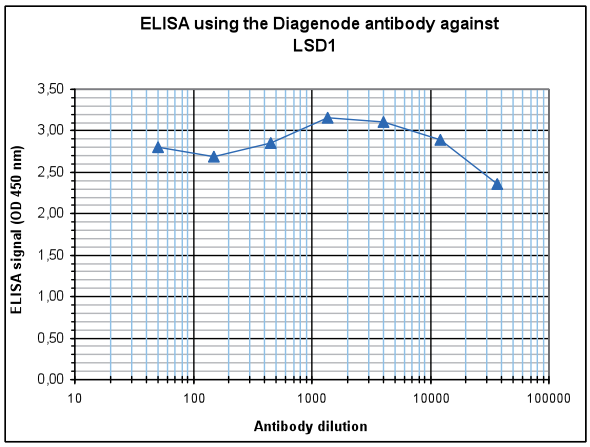

Figure 3. Determination of the antibody titer

To determine the titer of the antibody, an ELISA was performed using a serial dilution of the Diagenode antibody directed against LSD1 (Cat. No. C15410067) in antigen coated wells. By plotting the absorbance against the antibody dilution (Figure 3), the titer of the antibody was estimated to be 1:176,000.

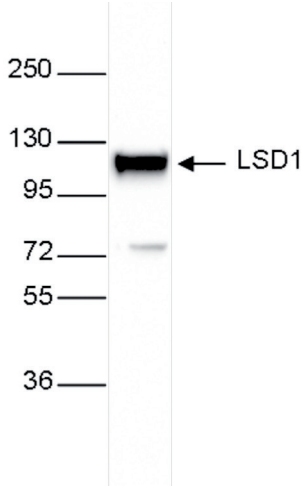

Figure 4. Western blot analysis using the Diagenode antibody directed against LSD1

Western blot was performed using nuclear extracts from HeLa cells (40 µg) and the Diagenode antibody against LSD1 (Cat. No. C15410067) diluted 1:4,000 in TBS-Tween containing 5% skimmed milk. The molecular weight marker (in kDa) is shown on the left. The location of the protein of interest is indicated on the right.

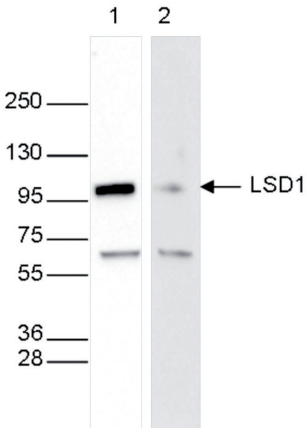

Figure 5. Western blot analysis using the Diagenode antibody directed against LSD1

Whole cell extracts (40 µg) from HeLa cells transfected with LSD1 siRNA (lane 2) and from an untransfected control (lane 1) were analysed by Western blot using the Diagenode antibody against LSD1 (Cat. No. C15410067) diluted 1:5,000 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.

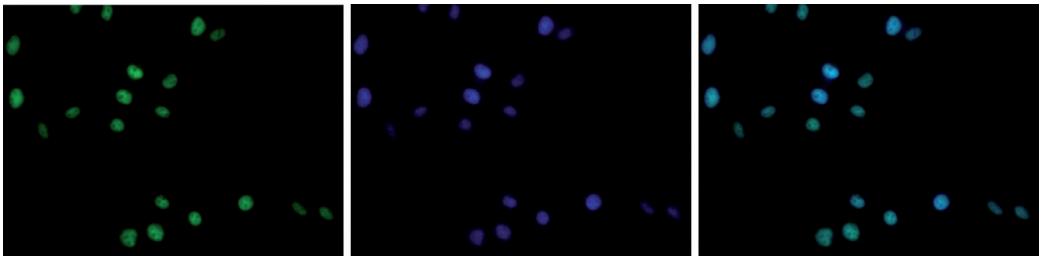

Figure 6. Immunofluorescence using the Diagenode antibody directed against LSD1

HeLa cells were stained with the Diagenode antibody against LSD1 (Cat. No. C15410067) 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 labelled with the LSD1 antibody (left) diluted 1:200 in blocking solution followed by an anti-rabbit antibody conjugated to Alexa488. The middle 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

Last update: November 2, 2015