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Ref-1 (h): 293T Lysate: sc-176689

BACKGROUND

The role of transcription factors in the regulation of gene expression is well established. Although the activity of these factors can be regulated by phosphorylation, evidence has indicated regulation of DNA binding mediated by changes in reduction-oxidation (redox) status. Mutational analysis has identified a single conserved cysteine residue mapping within the DNA binding domains of Fos and Jun. Chemical oxidation or modification of this cysteine residue inhibits the DNA binding activity of Fos and Jun. A similar mode of regulation has been recently proposed for other nuclear transcription factors. Oxidation is reversible by these compounds or by a cellular redox/DNA repair protein identified originally as Ref-1 (redox factor 1). Ref-1 is identical to a previously characterized DNA repair enzyme designated HAP1, APE or APEX.

REFERENCES

1. Abate, C., et al. 1990. Redox regulation of Fos and Jun DNA binding activity *in vitro*. *Science* 249: 1157-1161.
2. Boyle, W.J., et al. 1991. Activation of PKC decreases phosphorylation of c-Jun at sites that only regulate its DNA binding activity. *Cell* 64: 573-584.
3. Hunter, T. and Karin, M. 1992. The regulation of transcription by phosphorylation. *Cell* 70: 375-387.
4. Guehmann, S., et al. 1992. Reduction of a conserved Cys is essential for Myb DNA-binding. *Nucleic Acids Res.* 20: 2279-2286.
5. Xanthoudakis, S. and Curran, T. 1992. Identification and characterization of Ref-1, a nuclear protein that facilitates AP-1 DNA-binding activity. *EMBO J.* 11: 653-665.
6. Xanthoudakis, S., et al. 1992. Redox activation of Fos-Jun DNA binding activity is mediated by a DNA repair enzyme. *EMBO J.* 11: 3323-3335.
7. Walker, L.J., et al. 1993. Identification of residues in the human DNA repair enzyme HAP1 (Ref-1) that are essential for redox regulation of Jun DNA binding. *Mol. Cell. Biol.* 13: 5370-5376.
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CHROMOSOMAL LOCATION

Genetic locus: APEX1 (human) mapping to 14q11.2.

PRODUCT

Ref-1 (h): 293T Lysate represents a lysate of human Ref-1 transfected 293T cells and is provided as 100 µg protein in 200 µl SDS-PAGE buffer.

APPLICATIONS

Ref-1 (h): 293T Lysate is suitable as a Western Blotting positive control for human reactive Ref-1 antibodies. Recommended use: 10-20 µl per lane.

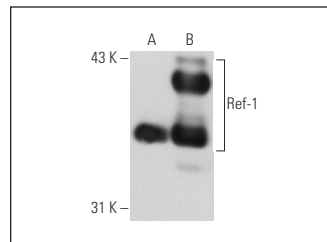
Control 293T Lysate: sc-117752 is available as a Western Blotting negative control lysate derived from non-transfected 293T cells.

Ref-1 (C-4): sc-17774 is recommended as a positive control antibody for Western Blot analysis of enhanced human Ref-1 expression in Ref-1 transfected 293T cells (starting dilution 1:100, dilution range 1:100-1:1,000).

RECOMMENDED SUPPORT REAGENTS

To ensure optimal results, the following support reagents are recommended:
 1) Western Blotting: use m-IgGκ BP-HRP: sc-516102 or m-IgGκ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™ Molecular Weight Standards: sc-2035, UltraCruz® Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048.

DATA



Ref-1 (C-4): sc-17774. Western blot analysis of Ref-1 expression in non-transfected: sc-117752 (A) and human Ref-1 transfected: sc-176689 (B) 293T whole cell lysates.

STORAGE

Store at -20° C. Repeated freezing and thawing should be minimized. Sample vial should be boiled once prior to use. Non-hazardous. No MSDS required.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.