

FIH-1 (D-12): sc-365016

BACKGROUND

Factor inhibiting HIF-1 (FIH-1) exists as a homodimer and binds to HIF-1 α . Specifically, FIH-1 operates as an asparaginyl hydroxylase. FIH-1 catalyzes the hydroxylation of the β -carbon of asparagine residue 803 within the C-terminal transactivation domain of HIF-1 α . This hydroxylation event blocks the association of HIF-1 α with coactivators. FIH-1 also binds to von Hippel-Lindau (VHL) tumor suppressor protein, which represses transcriptional activity of HIF-1 α . In transiently transfected human osteosarcoma cells, FIH-1 localizes to the cytoplasm. The structure of FIH-1 includes a jellyroll-like β -barrel containing ferrous-binding triad residues. The gene encoding human FIH-1 maps to chromosome 10q24.31.

REFERENCES

1. Mahon, P.C., Hirota, K. and Semenza, G.L. 2001. FIH-1: a novel protein that interacts with HIF-1 α and VHL to mediate repression of HIF-1 transcriptional activity. *Genes Dev.* 15: 2675-2686.
2. Dann, C.E., 3rd, Bruick, R.K. and Deisenhofer, J. 2002. Structure of factor-inhibiting hypoxia-inducible factor 1: an asparaginyl hydroxylase involved in the hypoxic response pathway. *Proc. Natl. Acad. Sci. USA* 99: 15351-15356.
3. Lando, D., Peet, D.J., Gorman, J.J., Whelan, D.A., Whitelaw, M.L. and Bruick, R.K. 2002. FIH-1 is an asparaginyl hydroxylase enzyme that regulates the transcriptional activity of hypoxia-inducible factor. *Genes Dev.* 16: 1466-1471.
4. McNeill, L.A., Hewitson, K.S., Claridge, T.D., Seibel, J.F., Horsfall, L.E. and Schofield, C.J. 2002. Hypoxia-inducible factor asparaginyl hydroxylase (FIH-1) catalyses hydroxylation at the β -carbon of asparagine-803. *Biochem. J.* 367: 571-575.
5. Metzén, E., Berchner-Pfannschmidt, U., Stengel, P., Marxsen, J.H., Stolze, I., Klinger, M., Huang, W.Q., Wotzlaw, C., Hellwig-Burgel, T., Jelkmann, W., Acker, H. and Fandrey, J. 2003. Intracellular localisation of human HIF-1 α hydroxylases: implications for oxygen sensing. *J. Cell Sci.* 116: 1319-1326.

CHROMOSOMAL LOCATION

Genetic locus: HIF1AN (human) mapping to 10q24.31; Hif1an (mouse) mapping to 19 C3.

SOURCE

FIH-1 (D-12) is a mouse monoclonal antibody raised against amino acids 15-243 mapping within an internal region of FIH-1 of human origin.

PRODUCT

Each vial contains 200 μ g IgG₁ kappa light chain in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

STORAGE

Store at 4° C, **DO NOT FREEZE**. Stable for one year from the date of shipment. Non-hazardous. No MSDS required.

APPLICATIONS

FIH-1 (D-12) is recommended for detection of FIH-1 of mouse, rat and human origin by Western Blotting (starting dilution 1:100, dilution range 1:100-1:1000), immunoprecipitation [1-2 μ g per 100-500 μ g of total protein (1 ml of cell lysate)], immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500) and solid phase ELISA (starting dilution 1:30, dilution range 1:30-1:3000).

Suitable for use as control antibody for FIH-1 siRNA (h): sc-37885, FIH-1 siRNA (m): sc-37886, FIH-1 shRNA Plasmid (h): sc-37885-SH, FIH-1 shRNA Plasmid (m): sc-37886-SH, FIH-1 shRNA (h) Lentiviral Particles: sc-37885-V and FIH-1 shRNA (m) Lentiviral Particles: sc-37886-V.

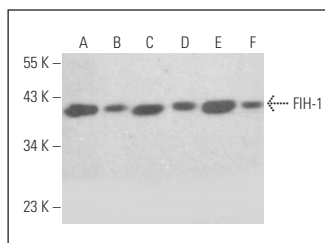
Molecular Weight of FIH-1: 40 kDa.

Positive Controls: HL-60 whole cell lysate: sc-2209, Jurkat whole cell lysate: sc-2204 or A-673 cell lysate: sc-2414.

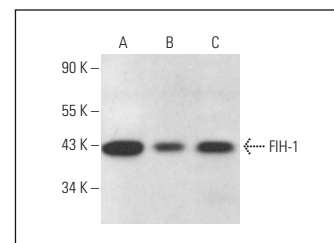
RECOMMENDED SUPPORT REAGENTS

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-IgG κ BP-HRP: sc-516102 or m-IgG κ 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. 2) Immunoprecipitation: use Protein A/G PLUS-Agarose: sc-2003 (0.5 ml agarose/2.0 ml). 3) Immunofluorescence: use m-IgG κ BP-FITC: sc-516140 or m-IgG κ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

DATA



FIH-1 (D-12): sc-365016. Western blot analysis of FIH-1 expression in Jurkat (A), A-375 (B), HL-60 (C), HEK293 (D), Raji (E) and A-431 (F) whole cell lysates.



FIH-1 (D-12): sc-365016. Western blot analysis of FIH-1 expression in Jurkat (A), MOLT-4 (B) and A-673 (C) whole cell lysates.

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.