

FOXD1 siRNA (h): sc-60649

BACKGROUND

The FOX family of transcription factors share a common DNA binding domain termed a winged-helix or forkhead domain. Many FOX proteins play important roles in development, metabolism, cancer and aging. FOXD1 (also designated brain factor 2 or BF-2) is involved in regulating inflammation as well as kidney and retinal development. FOXD1 regulates the activity of NFAT and NFκB. Deficiency of FOXD1 results in multiorgan, systemic inflammation, exaggerated T cell-derived cytokine production and T cell proliferation in autologous MLRs. In kidneys, FOXD1 controls the production of signals required for the normal transition of induced mesenchyme into tubular epithelium and full growth and branching of the collecting system. Deletion of FOXD1 results in renal abnormalities. FOXD2 acts as a modulator of T cell activation.

REFERENCES

1. Hatini, V., et al. 1996. Essential role of stromal mesenchyme in kidney morphogenesis revealed by targeted disruption of winged helix transcription factor BF-2. *Genes Dev.* 10: 1467-1478.
2. Dahle, M.K., et al. 2002. Mechanisms of FOXC2- and FOXD1-mediated regulation of the RIα subunit of cAMP-dependent protein kinase include release of transcriptional repression and activation by protein kinase B α and cAMP. *J. Biol. Chem.* 277: 22902-22908.
3. Zhang, H., et al. 2003. Transcriptional activation of placental growth factor by the forkhead/winged helix transcription factor FOXD1. *Curr. Biol.* 13: 1625-1629.
4. Johansson, C.C., et al. 2003. A winged helix forkhead (FOXD2) tunes sensitivity to cAMP in T lymphocytes through regulation of cAMP-dependent protein kinase RIα. *J. Biol. Chem.* 278: 17573-17579.

CHROMOSOMAL LOCATION

Genetic locus: FOXD1 (human) mapping to 5q13.2.

PRODUCT

FOXD1 siRNA (h) is a pool of 3 target-specific 19-25 nt siRNAs designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10 μM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see FOXD1 shRNA Plasmid (h): sc-60649-SH and FOXD1 shRNA (h) Lentiviral Particles: sc-60649-V as alternate gene silencing products.

For independent verification of FOXD1 (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-60649A, sc-60649B and sc-60649C.

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20° C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20° C, avoid contact with RNases and repeated freeze thaw cycles.

Resuspend lyophilized siRNA duplex in 330 μl of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μl of RNase-free water makes a 10 μM solution in a 10 μM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

FOXD1 siRNA (h) is recommended for the inhibition of FOXD1 expression in human cells.

SUPPORT REAGENTS

For optimal siRNA transfection efficiency, Santa Cruz Biotechnology's siRNA Transfection Reagent: sc-29528 (0.3 ml), siRNA Transfection Medium: sc-36868 (20 ml) and siRNA Dilution Buffer: sc-29527 (1.5 ml) are recommended. Control siRNAs or Fluorescein Conjugated Control siRNAs are available as 10 μM in 66 μl. Each contain a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA. Fluorescein Conjugated Control siRNAs include: sc-36869, sc-44239, sc-44240 and sc-44241. Control siRNAs include: sc-37007, sc-44230, sc-44231, sc-44232, sc-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

GENE EXPRESSION MONITORING

FOXD1 (2C10): sc-293238 is recommended as a control antibody for monitoring of FOXD1 gene expression knockdown by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500).

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. 2) Immunofluorescence: use m-IgGκ BP-FITC: sc-516140 or m-IgGκ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor FOXD1 gene expression knockdown using RT-PCR Primer: FOXD1 (h)-PR: sc-60649-PR (20 μl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Zhao, Y.F., et al. 2015. FOXD1 promotes breast cancer proliferation and chemotherapeutic drug resistance by targeting p27. *Biochem. Biophys. Res. Commun.* 456: 232-237.
2. Gao, Y.F., et al. 2017. Silencing of forkhead box D1 inhibits proliferation and migration in glioma cells. *Oncol. Rep.* 37: 1196-1202.

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.