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SZABO-SCANDIC HandelsgmbH

Quellenstraße 110, A-1100 Wien

T. +43(0)1 489 3961-0

F. +43(0)1 489 3961-7

mail@szabo-scandic.com

www.szabo-scandic.com

[linkedin.com/company/szaboscandic](https://www.linkedin.com/company/szaboscandic) 

FOXC2 shRNA (m) Lentiviral Particles: sc-45366-V

BACKGROUND

FOXC2 is a member of forkhead/winged helix transcription factor family, whose members serve as key regulators in embryogenesis and cell differentiation. FOXC2 functions as a key regulator of adipocyte metabolism by increasing the sensitivity of the β -adrenergic cAMP protein kinase A (PKA) signaling pathway through alteration of adipocyte PKA holoenzyme composition. Increased FOXC2 levels, induced by high fat diet, seem to counteract most of the symptoms associated with obesity. FOXC2 expression is also associated with the early stage of chondrogenic differentiation both *in vivo* and *in vitro*. FOXC2 haploinsufficiency results in lymphedema-distichiasis (LD), an autosomal dominant disorder that classically presents as lymphedema of the limbs and double rows of eyelashes (distichiasis). Mutant mice null for FOXC2 show defects in axial and cranial skeletogenesis, suggesting a requirement of FOXC2 for skeletal tissue development. FOXC2 interacts with FOXC1 in the Notch signaling pathway and in kidney and heart development.

REFERENCES

1. Fang, J., et al. 2000. Mutations in FOXC2 (MFH-1), a forkhead family transcription factor, are responsible for the hereditary lymphedema-distichiasis syndrome. *Am. J. Hum. Genet.* 67: 1382-1388.
2. Kume, T., et al. 2000. Murine forkhead/winged helix genes *Foxc1* (Mf1) and *Foxc2* (Mfh1) are required for the early organogenesis of the kidney and urinary tract. *Development* 127: 1387-1395.
3. Kume, T., et al. 2001. The murine winged helix transcription factors, FOXC1 and FOXC2, are both required for cardiovascular development and somitogenesis. *Genes Dev.* 15: 2470-2482.
4. Nifuji, A., et al. 2001. Bone morphogenetic protein regulation of forkhead/winged helix transcription factor *Foxc2* (Mfh1) in a murine mesodermal cell line C1 and in skeletal precursor cells. *J. Bone Miner. Res.* 16: 1765-1771.
5. Cederberg, A., et al. 2001. FOXC2 is a winged helix gene that counteracts obesity, hypertriglyceridemia, and diet-induced insulin resistance. *Cell* 106: 563-573.

CHROMOSOMAL LOCATION

Genetic locus: *Foxc2* (mouse) mapping to 8 E1.

PRODUCT

FOXC2 shRNA (m) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 3 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 μ l frozen stock containing 1.0×10^6 infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see FOXC2 siRNA (m): sc-45366 and FOXC2 shRNA Plasmid (m): sc-45366-SH as alternate gene silencing products.

STORAGE

Store lentiviral particles at -80° C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4° C for up to one week. Avoid repeated freeze thaw cycles.

APPLICATIONS

FOXC2 shRNA (m) Lentiviral Particles is recommended for the inhibition of FOXC2 expression in mouse cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μ l frozen viral stock containing 1.0×10^6 infectious units of virus (IFU); contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

GENE EXPRESSION MONITORING

FOXC2 (G-7): sc-515234 is recommended as a control antibody for monitoring of FOXC2 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker[™] compatible goat anti-mouse IgG-HRP: sc-2031 (dilution range: 1:2000-1:5000), Cruz Marker[™] Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz[™] Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor FOXC2 gene expression knockdown using RT-PCR Primer: FOXC2 (m)-PR: sc-45366-PR (20 μ l, 576 bp). Annealing temperature for the primers should be $55-60^\circ$ C and the extension temperature should be $68-72^\circ$ C.

BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.

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