



# FKHRL1 siRNA (h): sc-37887

## BACKGROUND

FKHRL1 (forkhead in rhabdomyosarcoma-like 1), also known as FOXO3 (forkhead box O3) or FOXO3A, is a 673 amino acid transcriptional activator that belongs to the FKHR subfamily of forkhead transcription factors. Transcriptional activation of FKHR proteins is regulated by the serine/threonine kinase Akt1, which phosphorylates FKHRL1 at Threonine 32 and Serine 253. Phosphorylation by Akt1 negatively regulates FKHRL1 by promoting its export from the nucleus. Phosphorylated FKHRL1 associates with 14-3-3 proteins and this complex is retained in the cytoplasm. Growth factor withdrawal stimulates FKHRL1 dephosphorylation and nuclear translocation, leading to FKHR-induced gene-specific transcriptional activation. Within the nucleus, dephosphorylated FKHRL1 triggers apoptosis by inducing the expression of genes that are critical for cell death.

## CHROMOSOMAL LOCATION

Genetic locus: FOXO3 (human) mapping to 6q21.

## PRODUCT

FKHRL1 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see FKHRL1 shRNA Plasmid (h): sc-37887-SH and FKHRL1 shRNA (h) Lentiviral Particles: sc-37887-V as alternate gene silencing products.

For independent verification of FKHRL1 (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-37887A, sc-37887B and sc-37887C.

## 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  $\mu$ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330  $\mu$ l of RNase-free water makes a 10  $\mu$ M solution in a 10  $\mu$ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

## APPLICATIONS

FKHRL1 siRNA (h) is recommended for the inhibition of FKHRL1 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  $\mu$ M in 66  $\mu$ l. 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

FKHRL1 (D-12): sc-48348 is recommended as a control antibody for monitoring of FKHRL1 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).

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor FKHRL1 gene expression knockdown using RT-PCR Primer: FKHRL1 (h) -PR: sc-37887-PR (20  $\mu$ l, 444 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

## SELECT PRODUCT CITATIONS

1. Stan, S.D., et al. 2008. Withaferin A causes FOXO3a- and Bim-dependent apoptosis and inhibits growth of human breast cancer cells *in vivo*. *Cancer Res.* 68: 7661-7669.
2. Guo, L., et al. 2012. Autophagy in premature senescent cells is activated via AMPK pathway. *Int. J. Mol. Sci.* 13: 3563-3582.
3. Liang, C., et al. 2013. Serotonin promotes the proliferation of serum-deprived hepatocellular carcinoma cells via upregulation of FOXO3a. *Mol. Cancer* 12: 14.
4. Hyun, S.W. and Jung, Y.S. 2014. Hypoxia induces FoxO3a-mediated dysfunction of blood-brain barrier. *Biochem. Biophys. Res. Commun.* 450: 1638-1642.
5. Ananda Sadagopan, S.K., et al. 2015. Forkhead box transcription factor (FOXO3a) mediates the cytotoxic effect of vernodalin *in vitro* and inhibits the breast tumor growth *in vivo*. *J. Exp. Clin. Cancer Res.* 34: 147.
6. Park, S.H., et al. 2016. Pharmacological activation of FOXO3 suppresses triple-negative breast cancer *in vitro* and *in vivo*. *Oncotarget* 7: 42110-42125.
7. Laporte, A.N., et al. 2017. Death by HDAC inhibition in synovial sarcoma cells. *Mol. Cancer Ther.* 16: 2656-2667.
8. Yue, D. and Sun, X. 2018. Idelalisib promotes Bim-dependent apoptosis through Akt/FoxO3a in hepatocellular carcinoma. *Cell Death Dis.* 9: 935.
9. Qian, F., et al. 2019. ING4 suppresses hepatocellular carcinoma via a NF- $\kappa$ B/miR-155/FOXO3a signaling axis. *Int. J. Biol. Sci.* 15: 369-385.
10. Li, J.R., et al. 2020. Endoplasmic reticulum stress and autophagy contributed to cadmium nephrotoxicity in HK-2 cells and sprague-dawley rats. *Food Chem. Toxicol.* 146: 111828.
11. Bagam, P., et al. 2021. *In vitro* study of the role of FOXO transcription factors in regulating cigarette smoke extract-induced autophagy. *Cell Biol. Toxicol.* 37: 531-553.

## RESEARCH USE

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