

SMRTe siRNA (h): sc-36514

BACKGROUND

Retinoids are metabolites of vitamin A (retinol) and represent important signaling molecules during vertebrate development and tissue differentiation. Retinoic acid receptors (RARs) have a high affinity for all *trans*-retinoic acids and belong to the same class of nuclear transcription factors as thyroid hormone receptors, vitamin D₃ receptor and ecdysone receptor. Two cofactors that function to repress transcription, designated SMRT (silencing mediator for RARs and thyroid receptors (TR)) and N-CoR, associate with TR and RAR in their unliganded state and are released from them upon ligand binding. The carboxy-termini of both proteins contain receptor interacting domains while their amino-termini contain two repressor domains. SMRT is comprised of 1,495 amino acids and contains an eight amino acid sequence that is not present in SMRTe (SMRT-extended), which contains 2,514 amino acids. SMRTe contains an N-terminal sequence spanning over 1,000 amino acids that is not present in SMRT, but that shows significant similarity with N-CoR. SMRTe expression is regulated during cell cycle progression, suggesting a role for SMRTe in the regulation of cycle-specific gene expression in diverse signaling pathways.

CHROMOSOMAL LOCATION

Genetic locus: NCOR2 (human) mapping to 12q24.31.

PRODUCT

SMRTe 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see SMRTe shRNA Plasmid (h): sc-36514-SH and SMRTe shRNA (h) Lentiviral Particles: sc-36514-V as alternate gene silencing products.

For independent verification of SMRTe (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-36514A, sc-36514B and sc-36514C.

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

APPLICATIONS

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

PROTOCOLS

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

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 μ M in 66 μ 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

SMRTe (1542/H7): sc-13554 is recommended as a control antibody for monitoring of SMRTe 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-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) 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor SMRTe gene expression knockdown using RT-PCR Primer: SMRTe (h)-PR: sc-36514-PR (20 μ l, 470 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Trtkova, K., et al. 2010. Binding of AR to SMRT/N-CoR complex and its co-operation with PSA promoter in prostate cancer cells treated with natural histone deacetylase inhibitor NaB. *Neoplasma* 57: 406-414.
2. Wargon, V., et al. 2014. Progestin and antiprogestin responsiveness in breast cancer is driven by the PRA/PRB ratio via AIB1 or SMRT recruitment to the CCND1 and MYC promoters. *Int. J. Cancer* 136: 2680-2692.
3. Zhang, F., et al. 2018. Ligand activation of PPAR γ by ligustrazine suppresses pericyte functions of hepatic stellate cells via SMRT-mediated transrepression of HIF-1 α . *Theranostics* 8: 610-626.
4. Yang, L., et al. 2022. Elucidating the novel mechanism of ligustrazine in preventing postoperative peritoneal adhesion formation. *Oxid. Med. Cell. Longev.* 2022: 9226022.

RESEARCH USE

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