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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) 

BRD4 siRNA (h): sc-43639

BACKGROUND

Brd4 belongs to the BET family, a group of structurally related proteins containing two bromodomains. Through these two domains, Brd4 associates with mitotic chromosomes and its expression correlates with cell growth. Expression of Brd4 inhibits cell cycle progression from G₁ to S, due to binding to the largest subunit of replication factor C (RFC) to prevent DNA elongation. Altered Brd4 function correlates with poorly differentiated carcinoma, with aggressive phenotype and a highly lethal outcome.

CHROMOSOMAL LOCATION

Genetic locus: BRD4 (human) mapping to 19p13.12.

PRODUCT

BRD4 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 BRD4 shRNA Plasmid (h): sc-43639-SH and BRD4 shRNA (h) Lentiviral Particles: sc-43639-V as alternate gene silencing products.

For independent verification of BRD4 (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-43639A, sc-43639B and sc-43639C.

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

BRD4 siRNA (h) is recommended for the inhibition of BRD4 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 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.

PROTOCOLS

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

GENE EXPRESSION MONITORING

BRD4 (A-7): sc-518021 is recommended as a control antibody for monitoring of BRD4 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 BRD4 gene expression knockdown using RT-PCR Primer: BRD4 (h)-PR: sc-43639-PR (20 μ l, 598 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Yao, W., et al. 2015. The BET bromodomain inhibitor, JQ1, facilitates c-FLIP degradation and enhances TRAIL-induced apoptosis independent of BRD4 and c-Myc inhibition. *Oncotarget* 6: 34669-34679.
2. Li, G.Q., et al. 2016. Suppression of BRD4 inhibits human hepatocellular carcinoma by repressing MYC and enhancing BIM expression. *Oncotarget* 7: 2462-2474.
3. Qu, M., et al. 2018. BRD4 inhibitor JQ1 inhibits and reverses mechanical injury-induced corneal scarring. *Cell Death Discov.* 4: 5.
4. Nerlakanti, N., et al. 2018. Targeting the BRD4-HOXB13 coregulated transcriptional networks with bromodomain-kinase inhibitors to suppress metastatic castration-resistant prostate cancer. *Mol. Cancer Ther.* 17: 2796-2810.
5. Tan, X., et al. 2019. BET inhibitors potentiate chemotherapy and killing of SPOP-mutant colon cancer cells via induction of DR5. *Cancer Res.* 79: 1191-1203.
6. Li, S., et al. 2019. miRNA-302e attenuates inflammation in infantile pneumonia through the RelA/BRD4/NF κ B signaling pathway. *Int. J. Mol. Med.* 44: 47-56.

RESEARCH USE

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