SANTA CRUZ BIOTECHNOLOGY, INC.

DRAM siRNA (h): sc-96209

BACKGROUND

DRAM (damage-regulated autophagy modulator) is a multi-pass membrane protein that belongs to the TMEM77 family of proteins and localizes to the lysosome membrane. DRAM is a highly conserved protein across many species and contains six transmembrane domains and an endoplasmic reticulum (ER) signal peptide. Its expression is induced by both p53 and p73, and it acts as a key player that is required (but not sufficient) for p53-induced autophagy and apoptosis. Although its expression is also induced by p73, DRAM is dispensable for p73-mediated apoptosis. As is suggested by its lysosomal localization, DRAM may participate in the degradation of proteins or in trafficking through the secretory pathway. In addition, DRAM expression is downregulated in human cancers, implying a profound role for DRAM in tumor development.

CHROMOSOMAL LOCATION

Genetic locus: DRAM (human) mapping to 12q23.2.

PRODUCT

DRAM 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 DRAM shRNA Plasmid (h): sc-96209-SH and DRAM shRNA (h) Lentiviral Particles: sc-96209-V as alternate gene silencing products.

For independent verification of DRAM (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-96209A, sc-96209B and sc-96209C.

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

DRAM siRNA (h) is recommended for the inhibition of DRAM 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

DRAM (M3-P4B4): sc-81713 is recommended as a control antibody for monitoring of DRAM 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-IgGλ BP-HRP: sc-516132 or m-IgGλ BP-HRP (Cruz Marker): sc-516132-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-IgGλ BP-FITC: sc-516185 or m-IgGλ BP-PE: sc-516186 (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 DRAM gene expression knockdown using RT-PCR Primer: DRAM (h)-PR: sc-96209-PR (20 µl, 576 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Maiuri, M.C., et al. 2009. Stimulation of autophagy by the p53 target gene Sestrin2. Cell Cycle 8: 1571-1576.
2. Hung, T.H., et al. 2012. Increased autophagy in placentas of intrauterine growth-restricted pregnancies. PLoS ONE 7: e40957.
3. Hung, T.H., et al. 2013. Autophagy in the human placenta throughout gestation. PLoS ONE 8: e83475.
4. Garufi, A., et al. 2017. p53-dependent PUMA to DRAM antagonistic interplay as a key molecular switch in cell-fate decision in normal/high glucose conditions. J. Exp. Clin. Cancer Res. 36: 126.
5. Meng, C., et al. 2018. MicroRNA-26b suppresses autophagy in breast cancer cells by targeting DRAM1 mRNA, and is downregulated by irradiation. Oncol. Lett. 15: 1435-1440.
6. Liu, D., et al. 2018. DNA damage regulated autophagy modulator 1 recovers the function of apoptosis-stimulating of p53 protein 2 on inducing apoptotic cell death in Huh7.5 cells. Oncol. Lett. 15: 9333-9338.
7. Kim, H.S., et al. 2022. The role of retinoid-related orphan receptor-α in cigarette smoke-induced autophagic response. Respir. Res. 23: 110.

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.