SANTA CRUZ BIOTECHNOLOGY, INC.

RNase III Drosha siRNA (h): sc-44080

BACKGROUND

The ribonuclease III superfamily represents a structurally distinct group of double-strand-specific endonucleases with essential roles in RNA maturation, RNA decay and gene silencing. Initial cleavage of microRNAs is catalysed by Drosha, a nuclease of the RNase III family, which acts on primary miRNA transcripts (pri-miRNAs) in the nucleus. Human Drosha is a component of two multi-protein complexes. The larger complex contains multiple classes of RNA-associated proteins including RNA helicases, proteins that bind double-stranded RNA, novel heterogeneous nuclear ribonucleoproteins and the Ewing's sarcoma family of proteins. The smaller complex is composed of Drosha and the double-stranded-RNA-binding protein, DGCR8.

REFERENCES

1. Denli A.M., et al. 2004. Processing of primary microRNAs by the microprocessor complex. Nature 432: 231-235.
2. Gregory R.I., et al. 2004. The microprocessor complex mediates the genesis of microRNAs. Nature 432: 235-240.
3. Sun W., et al. 2004. Mutational analysis of the nuclease domain of *Escherichia coli* ribonuclease III. Identification of conserved acidic residues that are important for catalytic function *in vitro.* Biochemistry 43: 13054-13062.

CHROMOSOMAL LOCATION

Genetic locus: DROSHA (human) mapping to 5p13.3.

PRODUCT

RNase III Drosha 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 RNase III Drosha shRNA Plasmid (h): sc-44080-SH and RNase III Drosha shRNA (h) Lentiviral Particles: sc-44080-V as alternate gene silencing products.

For independent verification of RNase III Drosha (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-44080A, sc-44080B and sc-44080C.

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

RNase III Drosha siRNA (h) is recommended for the inhibition of RNase III Drosha 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

RNase III Drosha (C-7): sc-393591 is recommended as a control antibody for monitoring of RNase III Drosha 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 RNase III Drosha gene expression knockdown using RT-PCR Primer: RNase III Drosha (h)-PR: sc-44080-PR (20 µl, 458 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Zhou, R., et al. 2009. NFκB p65-dependent transactivation of miRNA genes following *Cryptosporidium parvum* infection stimulates epithelial cell immune responses. PLoS Pathog. 5: e1000681.
2. Pichiorri, F., et al. 2013. *In vivo* NCL targeting affects breast cancer aggressiveness through miRNA regulation. J. Exp. Med. 210: 951-968.
3. Martinez, I., et al. 2017. An Exportin-1-dependent microRNA biogenesis pathway during human cell quiescence. Proc. Natl. Acad. Sci. USA 114: E4961-E4970.
4. Zeng, P., et al. 2021. ERK1/2 inhibition reduces vascular calcification by activating miR-126-3p-DKK1/LRP6 pathway. Theranostics 11: 1129-1146.
5. Li, Y., et al. 2023. The ubiquitin-specific protease USP36 associates with the microprocessor complex and regulates miRNA biogenesis by SUMOylating DGCR8. Cancer Res. Commun. 3: 459-470.

RESEARCH USE

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