SANTA CRUZ BIOTECHNOLOGY, INC.

CFTR siRNA (h): sc-35054

BACKGROUND

CFTR, for cystic fibrosis transmembrane conductance regulator, is a cyclic adenosine monophosphate (cAMP)-regulated chloride channel protein. CFTR belongs to the MDR subfamily within the ATP-binding transport protein family. It has two transmembrane domains (TMDs), two nucleotide binding domains (NBDs) and one regulatory domain. Mutations of CFTR are associated with cystic fibrosis (CF), a disease characterized by chronic bronchopulmonary disease, elevated sweat electrolytes and insufficient pancreatic function. CFTR mutations can also result in congenital bilateral absence of vas deferens (CBAVD), a form of male sterility that a majority of male CF patients exhibit.

REFERENCES

1. Riordan, J.R., et al. 1989. Identification of the cystic fibrosis gene: cloning and characterization of complementary DNA. Science 245: 1066-1073.
2. Tsui, L.C. 1992. The spectrum of cystic fibrosis mutations. Trends Genet. 8: 392-398.
3. Gabriel, S.E., et al. 1993. CFTR and outward rectifying chloride channels are distinct proteins with a regulatory relationship. Nature 363: 263-268.
4. Hoof, T., et al. 1994. Cystic fibrosis-type mutational analysis in the ATP-binding cassette transporter signature of human P-glycoprotein Mdr-1. J. Biol. Chem. 269: 20575-20583.

CHROMOSOMAL LOCATION

Genetic locus: CFTR (human) mapping to 7q31.2.

PRODUCT

CFTR 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 CFTR shRNA Plasmid (h): sc-35054-SH and CFTR shRNA (h) Lentiviral Particles: sc-35054-V as alternate gene silencing products.

For independent verification of CFTR (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-35054A, sc-35054B and sc-35054C.

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

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

CFTR (A-3): sc-376683 is recommended as a control antibody for monitoring of CFTR 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 CFTR gene expression knockdown using RT-PCR Primer: CFTR (h)-PR: sc-35054-PR (20 µl, 313 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Antigny, F., et al. 2011. Transient receptor potential canonical channel 6 links Ca^{2+} mishandling to cystic fibrosis transmembrane conductance regulator channel dysfunction in cystic fibrosis. Am. J. Respir. Cell Mol. Biol. 44: 83-90.
2. Gauthier, T.W., et al. 2017. Impaired defenses of neonatal mouse alveolar macrophage with cftr deletion are modulated by glutathione and TGFβ1. Physiol. Rep. 5: e13086.
3. Li, W., et al. 2018. CFTR inhibits the invasion and growth of esophageal cancer cells by inhibiting the expression of NFκB. Cell Biol. Int. 42: 1680-1687.
4. Leir, S.H., et al. 2020. An organoid model to assay the role of CFTR in the human epididymis epithelium. Cell Tissue Res. 381: 327-336.

RESEARCH USE

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