SANTA CRUZ BIOTECHNOLOGY, INC.

α2ML1 siRNA (h): sc-95683

BACKGROUND

α-2-macroglobulin (α-2M) is a homotetrameric serum protein consisting of four identical subunits that form dimers through disulfide bonds. Initially, α-2M was characterized as a pan-proteinase inhibitor that was able to "bait" proteinases into cleaving specific peptide sequences on α-2M. This interaction induces a conformational change in α-2M, thus enabling it to "trap" the proteinase and further inhibit its activity. Subsequently, α-2M has been shown to function as a carrier protein and regulator of cytokines during inflammation. Circulating transforming growth factor β (TGFβ) in serum is primarily bound to α-2M, which renders TGFβ inactive. Mutations and deletions in the gene encoding α-2M are associated with an increased incidence of Alzheimer's disease (AD). α-2-macroglobulin-like protein 1 (α2ML1) is a related protein that is expressed in the epidermis and may play a role in keratinocyte differentiation.

REFERENCES

1. Barrett, A.J., et al. 1973. The interaction of α_2-macroglobulin with proteinases. Characteristics and specificity of the reaction, and a hypothesis concerning its molecular mechanism. Biochem. J. 133: 709-724.
2. Tsuchiya, Y., et al. 1987. Sequence analysis of the putative regulatory region of rat α_2-macroglobulin gene. Gene 57: 73-80.
3. Borth, W., et al. 1990. Binding of IL-1 β to α-macroglobulins and release by thioredoxin. J. Immunol. 145: 3747-3754.
4. Poller, W., et al. 1992. Cloning of the human α_2-macroglobulin gene and detection of mutations in two functional domains: the bait region and the thiolester site. Hum. Genet. 88: 313-319.
5. Webb, D.J., et al. 1998. Localization of the binding site for transforming growth factor-β in human α_2-macroglobulin to a 20-kDa peptide that also contains the bait region. J. Biol. Chem. 273: 13339-13346.
6. Blacker, D., et al. 1998. α_2-macroglobulin is genetically associated with Alzheimer disease. Nat. Genet. 19: 357-360.
7. Galliano, M.F., et al. 2006. A novel protease inhibitor of the α2-macroglobulin family expressed in the human epidermis. J. Biol. Chem. 281: 5780-5789.

CHROMOSOMAL LOCATION

Genetic locus: A2ML1 (human) mapping to 12p13.31.

PRODUCT

α2ML1 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 α2ML1 shRNA Plasmid (h): sc-95683-SH and α2ML1 shRNA (h) Lentiviral Particles: sc-95683-V as alternate gene silencing products.

For independent verification of α2ML1 (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-95683A, sc-95683B and sc-95683C.

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

α2ML1 siRNA (h) is recommended for the inhibition of α2ML1 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

α2ML1 (G-8): sc-393082 is recommended as a control antibody for monitoring of α2ML1 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 α2ML1 gene expression knockdown using RT-PCR Primer: α2ML1 (h)-PR: sc-95683-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

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.