

C4 siRNA (h): sc-42844

BACKGROUND

The complement component proteins C3, C4 and C5 are potent anaphylatoxins that are released during complement activation, a system of ligand-surface protein interactions specific to cells of hematopoietic lineage. These proteins belong to the α -2-Macroglobulin family, but retain distinctive features including an anaphylatoxin domain and a netrin (NTR) domain. The human C4 gene is polymorphic at two loci, C4A and C4B, mapping to chromosome 6p21.33. C4A expresses the Rodgers (Rg) blood group Ag, while C4B expresses the Chido (Ch) blood group Ag. C4 is expressed as a precursor that is cleaved into α , β and γ chains, all of which are non-identical cleavage products. The α chain of C4 may be cleaved to produce an acidic isotype, C4a, which reacts with amino groups, and a basic isotype, C4b, which reacts with hydroxyl groups. Deficiency in the C4 gene is associated with autoimmune or immune complex disorders, such as systemic lupus erythematosus.

REFERENCES

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- Rupert, K.L., et al. 2002. The molecular basis of complete complement C4A and C4B deficiencies in a systemic lupus erythematosus patient with homozygous C4A and C4B mutant genes. J. Immunol. 169: 1570-1578.

CHROMOSOMAL LOCATION

Genetic locus: C4A/C4B (human) mapping to 6p21.33.

PRODUCT

C4 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 C4 shRNA Plasmid (h): sc-42844-SH and C4 shRNA (h) Lentiviral Particles: sc-42844-V as alternate gene silencing products.

For independent verification of C4 (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-42844A, sc-42844B and sc-42844C.

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

C4 siRNA (h) is recommended for the inhibition of C4 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.

GENE EXPRESSION MONITORING

C4 α (C-2): sc-271181 is recommended as a control antibody for monitoring of C4 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 C4 gene expression knockdown using RT-PCR Primer: C4 (h)-PR: sc-42844-PR (20 μ l, 432 bp). 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.