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SZABO-SCANDIC HandelsgmbH

Quellenstraße 110, A-1100 Wien

T. +43(0)1 489 3961-0

F. +43(0)1 489 3961-7

mail@szabo-scandic.com

www.szabo-scandic.com

linkedin.com/company/szaboscandic

SANTA CRUZ BIOTECHNOLOGY, INC.

Cryopyrin siRNA (h): sc-45469

BACKGROUND

Cryopyrin interacts selectively with apoptosis-associated specklike protein containing a CARD domain (ASC). This complex may function as an upstream activator of NFκB signaling and caspase-1 activation. The complex also inhibits TNFα induced activation and nuclear translocation of RelA/NFκB p65. Mutations in Cryopyrin and Pyrin proteins are responsible for several autoinflammatory disorders in humans, including familial cold autoinflammatory syndrome (FCAS), Muckle-Wells syndrome (MWS) and chronic infantile neurologic cutaneous and articular syndrome (CINCA).

CHROMOSOMAL LOCATION

Genetic locus: NLRP3 (human) mapping to 1q44.

PRODUCT

Cryopyrin 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 Cryopyrin shRNA Plasmid (h): sc-45469-SH and Cryopyrin shRNA (h) Lentiviral Particles: sc-45469-V as alternate gene silencing products.

For independent verification of Cryopyrin (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-45469A, sc-45469B and sc-45469C.

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

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

RESEARCH USE

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

GENE EXPRESSION MONITORING

Cryopyrin (6F12): sc-134306 is recommended as a control antibody for monitoring of Cryopyrin 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 Cryopyrin gene expression knockdown using RT-PCR Primer: Cryopyrin (h)-PR: sc-45469-PR (20 µl, 513 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Cho, K.A., et al. 2012. IL-17 and IL-22 enhance skin inflammation by stimulating the secretion of IL-1β by keratinocytes via the ROS-NLRP3-caspase-1 pathway. Int. Immunol. 24: 147-158.
2. Thinwa, J., et al. 2014. Integrin-mediated first signal for inflammasome activation in intestinal epithelial cells. J. Immunol. 193: 1373-1382.
3. Nagamatsu, K., et al. 2015. Dysregulation of *Escherichia coli* α-hemolysin expression alters the course of acute and persistent urinary tract infection. Proc. Natl. Acad. Sci. USA 112: E871-E880.
4. Sokolowska, M., et al. 2015. Prostaglandin E2 inhibits NLRP3 inflammasome activation through EP4 receptor and intracellular cyclic AMP in human macrophages. J. Immunol. 194: 5472-5487.
5. Dong, J., et al. 2017. Interleukin-22 participates in the inflammatory process of vitiligo. Oncotarget 8: 109161-109174.
6. Chen, M.L., et al. 2017. Trimethylamine-N-oxide induces vascular inflammation by activating the NLRP3 inflammasome through the SIRT3-SOD2-mtROS signaling pathway. J. Am. Heart Assoc. 6 pii: e002238.
7. Hu, Q., et al. 2018. Dihydromyricetin inhibits NLRP3 inflammasome-dependent pyroptosis by activating the Nrf2 signaling pathway in vascular endothelial cells. Biofactors 44: 123-136.
8. Shi, C.S., et al. 2019. SARS-coronavirus open reading frame-8b triggers intracellular stress pathways and activates NLRP3 inflammasomes. Cell Death Discov. 5: 101.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.