SANTA CRUZ BIOTECHNOLOGY, INC.

RAG-2 siRNA (m): sc-36372

BACKGROUND

Immunoglobulin (Ig) and T cell receptors of B and T lymphocytes are encoded in multiple germ line DNA segments known as V, D and J, that are rearranged during lymphocyte development. V(D)J recombination is a site specific recombination event in vertebrate genes. The assembly of antigen receptor genes by V(D)J recombination is initiated by a recombination activator genes 1 and 2 (RAG1/RAG2) protein complex, which introduces double-strand breaks between recombination signal sequences and their coding DNA. The RAG-1 and RAG-2 were originally identified on the basis of their ability to activate rearrangement of an exogenous recombinational substrate in fibroblasts; moreover, both genes are required for this activity. RAG1 and RAG2 proteins catalyze V(D)J are essential for generation of the diverse repertoire of antigen receptor genes and effective immune responses. RAG2 is composed of a "core" domain that is required for the recombination reaction and a C-terminal nonessential or "non-core" region. Activated mature CD5-positive human tonsil B cells coexpress both RAG1 and RAG2 mRNA and protein, and display DNA cleavage resulting from their recombinase activity.

REFERENCES

1. Schatz, D.G., et al. 1989. The V(D)J recombination activating gene, RAG-1. Cell 59: 1035-1048.
2. Shinkai, Y., et al. 1992. RAG-2-deficient mice lack mature lymphocytes owing to inability to initiate V(D)J rearrangement. Cell 68: 855-867.
3. Mombaerts, P., et al. 1992. RAG-1-deficient mice have no mature B and T lymphocytes. Cell 68: 869-877.
4. Schatz, D.G., et al. 1992. V(D)J recombination: molecular biology and regulation. Annu. Rev. Immunol. 10: 359-383.
5. Lin, W. and Desiderio, S. 1993. Regulation of V(D)J recombination activator protein RAG-2 by phosphorylation. Science 260: 953-959.
6. Chen, J., et al. 1993. RAG-2-deficient blastocyst complementation: an assay of gene function in lymphocyte development. Proc. Natl. Acad. Sci. USA 90: 4528-4532.
7. Wang, L.C. and Rosenberg, N. 1993. RAG-1 and RAG-2 are not sufficient to direct all phases of immunoglobulin gene rearrangement in pre-B-cell lines. Mol. Cell. Biol. 13: 3890-3899.

CHROMOSOMAL LOCATION

Genetic locus: Rag2 (mouse) mapping to 2 E2.

PRODUCT

RAG-2 siRNA (m) 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 RAG-2 shRNA Plasmid (m): sc-36372-SH and RAG-2 shRNA (m) Lentiviral Particles: sc-36372-V as alternate gene silencing products.

For independent verification of RAG-2 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-36372A, sc-36372B and sc-36372C.

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

RAG-2 siRNA (m) is recommended for the inhibition of RAG-2 expression in mouse 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor RAG-2 gene expression knockdown using RT-PCR Primer: RAG-2 (m)-PR: sc-36372-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.