

adenosine deaminase siRNA (h): sc-29644

BACKGROUND

Adenosine deaminase is an enzyme that is present in most tissues. It exists predominantly as a monomer, although in some tissues it is associated with adenosine deaminase-binding protein. Adenosine deaminase degrades extracellular adenosine, which is toxic for lymphocytes. Adenosine deaminase also effects co-stimulatory signals in T cells via interactions with CD26. Deficiency of adenosine deaminase has been shown to lead to immunodeficiency diseases such as SCID (severe combined immunodeficiency disease) and has been associated with hereditary hemolytic anemia, a disease in which adenosine deaminase levels are elevated fifty to seventy fold.

REFERENCES

1. Daddona, P.E., et al. 1980. Analysis of normal and mutant forms of human adenosine deaminase—a review. *Mol. Cell. Biochem.* 29: 91-101.
2. Miwa, S., et al. 1996. Molecular basis of erythroenzymopathies associated with hereditary hemolytic anemia: tabulation of mutant enzymes. *Am. J. Hematol.* 51: 122-132.
3. Resta, R., et al. 1997. SCID: the role of adenosine deaminase deficiency. *Immunol. Today* 18: 371-374.
4. Dong, R.P., et al. 1997. Determination of adenosine deaminase binding domain on CD26 and its immunoregulatory effect on T cell activation. *J. Immunol.* 259: 6070-6076.
5. Franco, R., et al. 1998. Enzymatic and extraenzymatic role of ectoadenosine deaminase in lymphocytes. *Immunol. Rev.* 161: 27-42.

CHROMOSOMAL LOCATION

Genetic locus: ADA (human) mapping to 20q13.12.

PRODUCT

adenosine deaminase 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 adenosine deaminase shRNA Plasmid (h): sc-29644-SH and adenosine deaminase shRNA (h) Lentiviral Particles: sc-29644-V as alternate gene silencing products.

For independent verification of adenosine deaminase (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-29644A, sc-29644B and sc-29644C.

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

adenosine deaminase siRNA (h) is recommended for the inhibition of adenosine deaminase 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

adenosine deaminase (D-4): sc-28346 is recommended as a control antibody for monitoring of adenosine deaminase 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 adenosine deaminase gene expression knockdown using RT-PCR Primer: adenosine deaminase (h)-PR: sc-29644-PR (20 μ l, 567 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Nakajima, Y., et al. 2015. Adenosine deaminase inhibitor EHNA exhibits a potent anticancer effect against malignant pleural mesothelioma. *Cell. Physiol. Biochem.* 35: 51-60.

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.