SANTA CRUZ BIOTECHNOLOGY, INC.

ART3 siRNA (m): sc-72538

BACKGROUND

Mono-ADP-ribosylation is one of the posttranslational protein modifications regulating cellular metabolism (e.g. nitrogen fixation) in prokaryotes. Mono-ADP-ribosylation is a process in which the ADP-ribose moiety of nicotinamide adenine dinucleotide is transferred to an acceptor amino acid. Five mammalian ADP-ribosyltransferases (ART1-ART5) have been cloned and expression is restricted to tissues such as cardiac and skeletal muscle, leukocytes, brain and testis. ART3 (ADP-ribosyltransferase 3), also known as ecto-ADP-ribosyltransferase 3, is a testis specific membrane protein that does not appear to have ADP-ribosyltransferase activity. It lacks the R-S-EXE active site motif and is therefore unable to catalyze the reaction. ART3 is predominantly found in spermatocytes and may play a role in spermatogenesis.

REFERENCES

1. Okazaki, I.J., et al. 1994. Immunological and structural conservation of mammalian skeletal muscle glycosylphosphatidylinositol-linked ADP-ribosyltransferases. Biochemistry 33: 12828-12836.
2. Koch-Nolte, F., et al. 1997. Two novel human members of an emerging mammalian gene family related to mono-ADP-ribosylating bacterial toxins. Genomics 39: 370-376.
3. Braren, R., et al. 1998. Molecular characterization and expression of the gene for mouse NAD^+: arginine ecto-mono (ADP-ribosyl) transferase, ART1. Biochem. J. 336: 561-568.
4. Okazaki, I.J. and Moss, J. 1999. Characterization of glycosylphosphatidylinositiol-anchored, secreted, and intracellular vertebrate mono-ADP-ribosyltransferases. Annu. Rev. Nutr. 19: 485-509.
5. Koch-Nolte, F., et al. 2005. Use of genetic immunization to raise antibodies recognizing toxin-related cell surface ADP-ribosyltransferases in native conformation. Cell. Immunol. 236: 66-71.
6. Friedrich, M., et al. 2006. Genomic organization and expression of the human mono-ADP-ribosyltransferase ART3 gene. Biochim. Biophys. Acta 1759: 270-280.

CHROMOSOMAL LOCATION

Genetic locus: Art3 (mouse) mapping to 5 E2.

PRODUCT

ART3 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 ART3 shRNA Plasmid (m): sc-72538-SH and ART3 shRNA (m) Lentiviral Particles: sc-72538-V as alternate gene silencing products.

For independent verification of ART3 (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-72538A, sc-72538B and sc-72538C.

RESEARCH USE

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

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

ART3 siRNA (m) is recommended for the inhibition of ART3 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.

GENE EXPRESSION MONITORING

ART3 (G-10): sc-515157 is recommended as a control antibody for monitoring of ART3 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 ART3 gene expression knockdown using RT-PCR Primer: ART3 (m)-PR: sc-72538-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

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