SANTA CRUZ BIOTECHNOLOGY, INC.

ALG-2 siRNA (m): sc-141006

BACKGROUND

An increased intracellular Ca^{2+} concentration induces apoptotic cell death. Transiently elevated Ca^{2+} concentrations are required for glucocorticoid-mediated and T cell receptor-mediated pathways, leading to T cell apoptosis. ALG-2 (for apoptosis-linked gene 2) is a Ca^{2+}-binding protein that participates in regulatory events occuring late in the apoptotic program and where several death signals converge. ALG-2 is a protein expressed in normal brain, and to a greater extent in ischemic brain. The ALG-2 protein contains five EF-hand-like motifs and shares homology with members of the penta EF-hand family, which includes Calpain small subunits sorcin and Grancalcin.

REFERENCES

1. McConkey, D.J., et al. 1989. Calcium-dependent killing of immature thymocytes by stimulation via the CD3/T cell receptor complex. J. Immunol. 143: 1801-1806.
2. McConkey, D.J., et al. 1989. Glucocorticoids activate a suicide process in thymocytes through an elevation of cytosolic Ca^{2+} concentration. Arch. Biochem. Biophys. 269: 365-370.
3. Nicotera, P., et al. 1990. The role of Ca^{2+} in cell killing. Chem. Res. Toxicol. 3: 484-494.
4. Vito, P., et al. 1996. Interfering with apoptosis: Ca^{2+}-binding protein ALG-2 and Alzheimer's disease gene ALG-3. Science 271: 521-525.
5. D'Adamio, L., et al. 1997. Functional cloning of genes involved in T cell receptor-induced programmed cell death. Semin. Immunol. 9: 17-23.
6. Maki, M., et al. 1997. A growing family of the Ca^{2+}-binding proteins with five EF-hand motifs. Biochem. J. 328: 718-720.
7. Venn, M.K. and Conway, E.L. 1998. Localization of mRNA for the apoptosis-linked gene ALG-2 in young and aged rat brain. Neuroreport 9: 1981-1985.
8. Li, W., et al. 2000. Increased expression of apoptosis-linked gene 2 (ALG-2) in the rat brain after temporary focal cerebral ischemia. Neuroscience 96: 161-168.

CHROMOSOMAL LOCATION

Genetic locus: Pdcd6 (mouse) mapping to 13 C1.

PRODUCT

ALG-2 siRNA (m) is a pool of 2 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 ALG-2 shRNA Plasmid (m): sc-141006-SH and ALG-2 shRNA (m) Lentiviral Particles: sc-141006-V as alternate gene silencing products.

For independent verification of ALG-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-141006A and sc-141006B.

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

ALG-2 siRNA (m) is recommended for the inhibition of ALG-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.

GENE EXPRESSION MONITORING

ALG-2 (H-11): sc-376950 is recommended as a control antibody for monitoring of ALG-2 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 ALG-2 gene expression knockdown using RT-PCR Primer: ALG-2 (m)-PR: sc-141006-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.