SANTA CRUZ BIOTECHNOLOGY, INC.

Sam50 siRNA (h): sc-76440

BACKGROUND

Sam50 (sorting assembly machinery component 50 homolog, *S. cerevisiae),* also known as SAMM50, OMP85, TOB55, TRG-3, CGI-51 or YNL026W, is a β-barrel multi-pass membrane protein that belongs to the SAM50/omp85 family of proteins. Localizing to the mitochondrion, Sam50 is believed to function in the assembly pathway of mitochondrial outer membrane β-barrel proteins. More specifically, Sam50 functions as the major component of the SAM (sorting and assembly machinery) complex, also known as the TOB (topogenesis of mitochondrial outer membrane β-barrel proteins) complex, and is required for cell viability. Exposed to the intermembrane space (IMS), the N-terminal POTRA (polypeptide transport-associated) domain of Sam50 (a domain that is conserved from bacteria to man) functions like a receptor for β-barrel proteins. The association of the Sam35 subunit of the SAM complex is essential for Sam50 binding to outer membrane substrate proteins.

REFERENCES

1. Kozjak, V., et al. 2003. An essential role of Sam50 in the protein sorting and assembly machinery of the mitochondrial outer membrane. J. Biol. Chem. 278: 48520-48523.
2. Meisinger, C., et al. 2004. The mitochondrial morphology protein Mdm10 functions in assembly of the preprotein translocase of the outer membrane. Dev. Cell 7: 61-71.
3. Humphries, A.D., et al. 2005. Dissection of the mitochondrial import and assembly pathway for human Tom40. J. Biol. Chem. 280: 11535-11543.
4. Habib, S.J., et al. 2005. Assembly of the TOB complex of mitochondria. J. Biol. Chem. 280: 6434-6440.
5. Paschen, S.A., et al. 2005. Biogenesis of β-barrel membrane proteins of mitochondria. Trends Biochem. Sci. 30: 575-582.
6. Meisinger, C., et al. 2006. Mitochondrial protein sorting: differentiation of β-barrel assembly by Tom7-mediated segregation of Mdm10. J. Biol. Chem. 281: 22819-22826.
7. Habib, S.J., et al. 2007. The N-terminal domain of Tob55 has a receptor-like function in the biogenesis of mitochondrial β-barrel proteins. J. Cell Biol. 176: 77-88.

CHROMOSOMAL LOCATION

Genetic locus: SAMM50 (human) mapping to 22q13.31.

PRODUCT

Sam50 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 Sam50 shRNA Plasmid (h): sc-76440-SH and Sam50 shRNA (h) Lentiviral Particles: sc-76440-V as alternate gene silencing products.

For independent verification of Sam50 (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-76440A, sc-76440B and sc-76440C.

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

Sam50 siRNA (h) is recommended for the inhibition of Sam50 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.

GENE EXPRESSION MONITORING

Sam50 (SQ-7): sc-100493 is recommended as a control antibody for monitoring of Sam50 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 Sam50 gene expression knockdown using RT-PCR Primer: Sam50 (h)-PR: sc-76440-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.