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GTPBP9 siRNA (m): sc-145833

BACKGROUND

GTP-binding protein 9 (GTPBP9), also known as Obg-like ATPase 1 (OLA1), is a 396 amino acid protein that belongs to the Obg-related GTPase family under the translation factors (TRAFAC) class. Originally thought to only have GTPase activity, Obg-related GTPase family members have been shown to also have ATPase activity. In *Homo sapiens*, GTPBP9 exhibits a preference for binding ATP over GTP, with GTP binding occurring only at high nucleotide concentration. One cause for ATP affinity and GTP discrimination is thought to be a substitution of glutamine for a hydrophobic amino acid in Obg-related family members; this is the same substitution that inactivates Ras-like GTPases. GTPBP9 contains a C-terminal TGS domain that binds to ligands and an N-terminal G domain which binds nucleotides. GTPBP9 is expressed as three isoforms produced by alternative splicing.

REFERENCES

1. Vetter, I.R. and Wittinghofer, A. 2001. The guanine nucleotide-binding switch in three dimensions. *Science* 294: 1299-1304.
2. Leipe, D.D., et al. 2002. Classification and evolution of P-loop GTPases and related ATPases. *J. Mol. Biol.* 317: 41-72.
3. Caldon, C.E. and March, P.E. 2003. Function of the universally conserved bacterial GTPases. *Curr. Opin. Microbiol.* 6: 135-139.
4. Teplyakov, A., et al. 2003. Crystal structure of the YchF protein reveals binding sites for GTP and nucleic acid. *J. Bacteriol.* 185: 4031-4037.
5. Brown, E.D. 2005. Conserved P-loop GTPases of unknown function in bacteria: an emerging and vital ensemble in bacterial physiology. *Biochem. Cell Biol.* 83: 738-746.

CHROMOSOMAL LOCATION

Genetic locus: Ola1 (mouse) mapping to 2 C3.

PRODUCT

GTPBP9 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see GTPBP9 shRNA Plasmid (m): sc-145833-SH and GTPBP9 shRNA (m) Lentiviral Particles: sc-145833-V as alternate gene silencing products.

For independent verification of GTPBP9 (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-145833A, sc-145833B and sc-145833C.

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

GTPBP9 siRNA (m) is recommended for the inhibition of GTPBP9 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 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

GTPBP9 (G-6): sc-393946 is recommended as a control antibody for monitoring of GTPBP9 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 GTPBP9 gene expression knockdown using RT-PCR Primer: GTPBP9 (m)-PR: sc-145833-PR (20 μ l, 513 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Martínez-Baeza, E., et al. 2016. Metal mixture (As-Cd-Pb)-induced cell transformation is modulated by OLA1. *Mutagenesis* 31: 463-473.
2. Xie, D., et al. 2020. OLA1 is responsible for normal spindle assembly and SAC activation in mouse oocytes. *PeerJ* 8: e8180.

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.