

Gab 3 siRNA (m): sc-40609

BACKGROUND

The Gab (GRB2-associated binder)/DOS ("Daughter of Sevenless") (Gab) family of adaptor proteins function as molecular scaffolds that mediate protein recruitment to RTKs. Cytokine/growth factor triggering of protein tyrosine kinase receptors (RTKs) initiates signaling cascades that progress to the nucleus where signals for activation, proliferation and differentiation occur. This scaffolding mechanism represents a critical link in cytokine/growth factor signaling routes. Gab 1-3 contain pleckstrin homology and potential binding sites for SH2 and SH3 domain-containing proteins. The recruitment of signaling partners to Gab family members is phosphorylation dependent. Insulin receptor and EGF-receptor signaling are among the cascades that rely on Gab family members to elicit a nuclear response to an extracellular stimulus. The human Gab 3 gene maps to chromosome Xq28 and encodes a 586 amino acid protein.

REFERENCES

1. Araki, E., et al. 1994. Alternative pathway of Insulin signaling in mice with targeted disruption of the IRS-1 gene. *Nature* 372: 186-190.
2. Holgado-Madruga, M., et al. 1996. A GRB2-associated docking protein in EGF- and Insulin-receptor signalling. *Nature* 379: 560-564.
3. Zhao, C., et al. 1999. Gab 2, a new pleckstrin homology domain-containing adapter protein, acts to uncouple signaling from ERK kinase to Elk-1. *J. Biol. Chem.* 274: 19649-19654.
4. Lock, L.S., et al. 2000. Identification of an atypical GRB2 carboxyl-terminal SH3 domain binding site in Gab docking proteins reveals GRB2-dependent and -independent recruitment of Gab 1 to receptor tyrosine kinases. *J. Biol. Chem.* 275: 31536-31545.
5. Wolf, I., et al. 2002. Gab 3, a new DOS/Gab family member, facilitates macrophage differentiation. *Mol. Cell. Biol.* 22: 231-244.
6. Online Mendelian Inheritance in Man, OMIM[™]. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 604439. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
7. LocusLink Report (LocusID: 139716). <http://www.ncbi.nlm.nih.gov/LocusLink/>

CHROMOSOMAL LOCATION

Genetic locus: Gab3 (mouse) mapping to X A7.3.

PRODUCT

Gab 3 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 Gab 3 shRNA Plasmid (m): sc-40609-SH and Gab 3 shRNA (m) Lentiviral Particles: sc-40609-V as alternate gene silencing products.

For independent verification of Gab 3 (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-40609A, sc-40609B and sc-40609C.

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

Gab 3 siRNA (m) is recommended for the inhibition of Gab 3 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

Gab 3 (F-1): sc-376456 is recommended as a control antibody for monitoring of Gab 3 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 Gab 3 gene expression knockdown using RT-PCR Primer: Gab 3 (m)-PR: sc-40609-PR (20 μ l). 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.