SANTA CRUZ BIOTECHNOLOGY, INC.

Gab 2 siRNA (m): sc-40607

BACKGROUND

Growth factor triggering of protein tyrosine kinase receptors induces signals that cascade to the nucleus, activating mitogenic as well as other responses. Critical components of this process include adapter protein such as Shc, IRS-1 and Gab 1 (GRB-associated binder-1) that lack detectable catalytic activity. These are immediate substrates of receptor tyrosine kinase activity and serve to link activated receptors to downstream signaling components. Whereas Shc has been implicated in signaling by diverse receptor families, IRS-1 serves primarily as the major Insulin receptor substrate. Shc and Gab 1 also participate in Insulin signaling by linking the Insulin receptor to Ras by forming complexes with GRB2 (another adapter protein) and Sos independently of IRS-1. The Gap 1 related protein, Gab 2, associates with SH2 domain-containing proteins, such as SHP2, and it is involved in a novel pathway for cytokine-induced gene activation.

REFERENCES

1. McGlade, J., et al. 1992. Shc proteins are phosphorylated and regulated by the v-Src and v-Fps protein-tyrosine kinase. Proc. Natl. Acad. Sci. USA 89: 8869-8873.
2. Pelicci, G., et al. 1992. A novel transforming protein (SHC) with an SH2 domain is implicated in mitogenic signal transduction. Cell 70: 93-104.
3. Lee, C.H., et al. 1993. Nck associates with the SH2 domain-docking protein IRS-1 in Insulin-stimulated cells. Proc. Natl. Acad. Sci. USA 90: 11713-11717.
4. Ravichandran, K.S., et al. 1993. Interaction of Shc with the ζ chain of the T cell receptor upon T cell activation. Science 262: 902-905.
5. Myers, M.G. et al. 1994. Role of IRS-1-GRB-2 complexes in Insulin signaling. Mol. Cell. Biol. 14: 3577-3587.
6. Tamemoto, K., et al. 1994. Insulin resistance and growth retardation in mice lacking Insulin receptor-substrate 1. Nature 372: 182-186.
7. Araki, E., et al. 1994. Alternative pathway of Insulin signaling in mice with targeted disruption of the IRS-1 gene. Nature 372: 186-190.
8. Gu, H., et al. 1998. Cloning of p97/Gab2, the major SHP2-binding protein in hematopoietic cells, reveals a novel pathway for cytokine-induced gene activation. Mol. Cell 2: 729-740.

CHROMOSOMAL LOCATION

Genetic locus: Gab2 (mouse) mapping to 7 E1.

PRODUCT

Gab 2 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 Gab 2 shRNA Plasmid (m): sc-40607-SH and Gab 2 shRNA (m) Lentiviral Particles: sc-40607-V as alternate gene silencing products.

For independent verification of Gab 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-40607A, sc-40607B and sc-40607C.

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

Gab 2 siRNA (m) is recommended for the inhibition of Gab 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

Gab 2 (H-6): sc-365590 is recommended as a control antibody for monitoring of Gab 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 Gab 2 gene expression knockdown using RT-PCR Primer: Gab 2 (m)-PR: sc-40607-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.