SANTA CRUZ BIOTECHNOLOGY, INC.

GRB10 siRNA (h): sc-35509

BACKGROUND

Many growth factors function by binding receptors with intrinsic tyrosine kinase activity. Signaling by such receptors involves a series of intermediates characterized by SH2 domains that bind tyrosine-phosphorylated receptors by a direct interaction between the SH2 domain and specific phospho-tyrosine-containing receptor sequences. GRB7, a SH2 domain protein, has a single SH2 domain at its C-terminal, a central region with similarity to Ras GAP and a proline-rich N terminus. A related SH2 domain-containing protein, GRB10, exhibits a high degree of homology with GRB7. GRB10 undergoes serine but not tyrosine phosphorylation in response to EGF treatment, but appears to bind to the EGF receptor poorly. GRB10 maps to mouse chromosome 11 A1, in close proximity to the EGF receptor. Similarly, GRB7 maps to the same mouse chromosome near the EGF receptor-related protein HER2.

REFERENCES

1. Schlessinger, J. et al. 1992. Growth factor signalling by receptor tyrosine kinases. Neuron 9: 383-391.
2. Margolis, B. 1992. Proteins with SH2 domains: transducers in the tyrosine kinase signalling pathway. Cell Growth Differ. 3: 73-80.
3. Margolis, B., et al. 1992. High-efficiency expression/cloning of epidermal growth factor-receptor-binding proteins with Src homology 2 domains. Proc. Natl. Acad. Sci. USA 89: 8894-8898.
4. Fanti, W.J., et al. 1993. Signalling by receptor tyrosine kinases. Annu. Rev. Biochem. 62: 453-481.
5. Stein, D., et al. 1994. The SH2 domain protein GRB7 is co-amplified, over-expressed and in a tight complex with HER2 in breast cancer. EMBO J. 13: 1331-1340.
6. Ooi, J., et al. 1995. The cloning of GRB10 reveals a new family of SH2 domain proteins. Oncogene 10: 1621-1630.
7. Wandless, T.J. 1996. SH2 domains: a question of independence. Curr. Biol. 6: 125-127.

CHROMOSOMAL LOCATION

Genetic locus: GRB10 (human) mapping to 7p12.1.

PRODUCT

GRB10 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 GRB10 shRNA Plasmid (h): sc-35509-SH and GRB10 shRNA (h) Lentiviral Particles: sc-35509-V as alternate gene silencing products.

For independent verification of GRB10 (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-35509A, sc-35509B and sc-35509C.

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

GRB10 siRNA (h) is recommended for the inhibition of GRB10 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

GRB10 (C-11): sc-74509 is recommended as a control antibody for monitoring of GRB10 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 GRB10 gene expression knockdown using RT-PCR Primer: GRB10 (h)-PR: sc-35509-PR (20 µl, 557 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Singh, M., et al. 2013. HuR inhibits apoptosis by amplifying Akt signaling through a positive feedback loop. J. Cell. Physiol. 228: 182-189.

RESEARCH USE

For research use only, not for use in diagnostic procedures.