

GCS- α -2 siRNA (h): sc-41012

BACKGROUND

Guanylate cyclases belong to the adenylyl cyclase class-4/guanylyl cyclase family. There are two forms of guanylate cyclase. The soluble form, known as GCS or sGC, act as receptors for nitric oxide (NO). The membrane-bound receptor form, known as GC, are peptide hormone receptors. GCS is a cGMP-synthesizing enzyme, which is the major receptor for the neurotransmitter nitric oxide. It plays a crucial role in smooth muscle contractility, platelet reactivity and neurotransmission. GCS is a heme-containing heterodimer, consisting of one α subunit and one β subunit. The heme moiety mediates NO activation, and this heme group also binds carbon monoxide (CO), which weakly stimulates the enzyme. Both NO and CO stimulation are enhanced by the allosteric activator 3-(5'-hydroxymethyl-2'-furyl)-benzyl-indazole, YC-1. YC-1 can also stimulate GCS in a NO-independent manner. Both α and β subunits are required for cGMP generation, and at least two isoforms exist for each subunit. Heterodimers consisting of α -1/ β -1 and α -2/ β -1 have been identified, and both display similar enzymatic activity. The distribution of the β -2 subunit seems to be much more restricted than the β -1 subunit, with predominant expression in kidney and liver.

REFERENCES

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2. Wedel, B., et al. 1995. Functional domains of soluble guanylyl cyclase. *J. Biol. Chem.* 270: 24871-24875.
3. Bellamy, T., et al. 2000. Rapid desensitization of the nitric oxide receptor, soluble guanylyl cyclase, underlies diversity of cellular cGMP responses. *Proc. Natl. Acad. Sci. USA* 97: 2928-2933.
4. Lee, Y., et al. 2000. Human recombinant soluble guanylyl cyclase: expression, purification and regulation. *Proc. Natl. Acad. Sci. USA* 97: 10763-10768.
5. Ibarra, C., et al. 2001. Regional and age-dependent expression of the nitric oxide receptor, soluble guanylyl cyclase, in the human brain. *Brain Res.* 907: 54-60.
6. Koblin, M., et al. 2001. Nitric oxide activates the β -2 subunit of soluble guanylyl cyclase in the absence of a second subunit. *J. Biol. Chem.* 276: 30737-30743.
7. Martin, E., et al. 2001. YC-1 activation of human soluble guanylyl cyclase has both heme-dependent and heme-independent components. *Proc. Natl. Acad. Sci. USA* 98: 12938-12942.
8. Gibb, B. and Garthwaite, J. 2001. Subunits of nitric oxide receptor, soluble guanylyl cyclase, expressed in rat brain. *Eur. J. Neurosci.* 13: 539-544.

CHROMOSOMAL LOCATION

Genetic locus: GUCY1A2 (human) mapping to 11q22.3.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.

PRODUCT

GCS- α -2 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see GCS- α -2 shRNA Plasmid (h): sc-41012-SH and GCS- α -2 shRNA (h) Lentiviral Particles: sc-41012-V as alternate gene silencing products.

For independent verification of GCS- α -2 (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-41012A, sc-41012B and sc-41012C.

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

GCS- α -2 siRNA (h) is recommended for the inhibition of GCS- α -2 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 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor GCS- α -2 gene expression knockdown using RT-PCR Primer: GCS- α -2 (h)-PR: sc-41012-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.