SANTA CRUZ BIOTECHNOLOGY, INC.

$G_{\alpha\ o}$ siRNA (m): sc-37256

SANTA CRUZ BIOTECHNOLOGY

The Power to Question

BACKGROUND

Heterotrimeric G proteins function to relay information from cell surface receptors to intracellular effectors. Each of a very broad range of receptors specifically detects an extracellular stimulus (a photon, pheromone, odorant, hormone or neurotransmitter) while the effectors (i.e., adenyl cyclase), which act to generate one or more intracellular messengers, are less numerous. In mammals, G protein α, β and γ polypeptides are encoded by at least 16, 4 and 7 genes, respectively. Most interest in G proteins has been focused on their α subunits, since these proteins bind and hydrolyze GTP and most obviously regulate the activity of the best studied effectors. Four distinct classes of G_α subunits have been identified; these include G_s, G_i, G_q and $G_{\alpha\ 12/13}$. The G_i class comprises all the known α subunits that are susceptible to pertussis toxin modifications, including $G_{\alpha\ i\text{-}1}$, $G_{\alpha\ i\text{-}2}$, $G_{\alpha\ i\text{-}3}$, $G_{\alpha\ o}$, $G_{\alpha\ t1}$, $G_{\alpha\ t2}$, $G_{\alpha\ z}$ and $G_{\alpha\ gust}$. Of these, the three $G_{\alpha\ i}$ subtypes function to open atrial potassium channels.

REFERENCES

1. Jones, D.T., et al. 1990. Biochemical characterization of three stimulatory GTP-binding proteins. The large and small forms of G_s and the olfactory-specific G protein, G_{olf}. J. Biol. Chem. 265: 2671-2676.
2. Simon, M.I., et al. 1991. Diversity of G proteins in signal transduction. Science 252: 802-808.
3. Cali, J.J., et al. 1992. Selective tissue distribution of G protein γ subunits, including a new form of the γ subunits identified by cDNA cloning. J. Biol. Chem. 267: 24023-24027.
4. McLaughlin, S.K., et al. 1992. Gustducin is a taste-cell-specific G protein closely related to the transducins. Nature 357: 563-569.

CHROMOSOMAL LOCATION

Genetic locus: Gnao1 (mouse) mapping to 8 C5.

PRODUCT

$G_{\alpha\ o}$ siRNA (m) is a pool of 2 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 $G_{\alpha\ o}$ shRNA Plasmid (m): sc-37256-SH and $G_{\alpha\ o}$ shRNA (m) Lentiviral Particles: sc-37256-V as alternate gene silencing products.

For independent verification of $G_{\alpha\ o}$ (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-37256A and sc-37256B.

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

$G_{\alpha\ o}$ siRNA (m) is recommended for the inhibition of $G_{\alpha\ o}$ 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

$G_{\alpha\ o}$ (A2): sc-13532 is recommended as a control antibody for monitoring of $G_{\alpha\ o}$ 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 $G_{\alpha\ o}$ gene expression knockdown using RT-PCR Primer: $G_{\alpha\ o}$ (m)-PR: sc-37256-PR (20 µl, 591 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Liu, X., et al. 2005. Rapid, Wnt-induced changes in GSK3β associations that regulate β-catenin stabilization are mediated by G_α proteins. Curr. Biol. 15: 1989-1997.
2. Gao, Y. and Wang, H.Y. 2007. Inositol pentakisphosphate mediates Wnt/β-catenin signaling. J. Biol. Chem. 282: 26490-26502.
3. Bikkavilli, R.K., et al. 2008. $G_{\alpha\ o}$ mediates Wnt-JNK signaling through dishevelled 1 and 3, RhoA family members, and MEKK 1 and 4 in mammalian cells. J. Cell Sci. 121: 234-245.
4. Bikkavilli, R.K., et al. 2008. p38 mitogen-activated protein kinase regulates canonical Wnt-β-catenin signaling by inactivation of GSK3β. J. Cell Sci. 121: 3598-3607.

RESEARCH USE

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