β_2 -AR siRNA (h): sc-39866



The Power to Question

BACKGROUND

Adrenergic receptors (ARs) (the term "adrenergic" reflects the alternative name for epinephrine, adrenaline) include four general types (α_1 , α_2 , β_1 and β_2) which are found in different target tissues and differ in their affinities and responses to various agonists and antagonists. cDNA clones have been isolated for all of the major AR subtypes and a number of closely related receptors have been identified by this approach. Each of the receptors have been shown to consist of single polypeptide chains which transverse the plasma membrane seven times, presumably forming a bundle of helices within the membrane. These transmembrane regions are hydrophobic and are interconnected by extracellular and intracellular hydrophilic loops. The coupling of ARs to specific intracellular effectors is mediated through diverse heterotrimeric G proteins and is regulated by G protein-coupled receptor kinases (GRKs), cAMP-dependent protein kinase A and protein kinase C directed phosphorylation. β_2 -adrenergic receptors bind cathecholamines (epinephrine, norepinephrine) and influence development, behavior, cardiac function, smooth muscle tone, and metabolism. β_2 -AR signaling complexes can contain C L-type calcium channel Ca(V)1.2, G protein, adenylyl cyclase, cAMP-dependent kinase, and PP2A phosphatase.

REFERENCES

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- Collins, S., et al. 1991. Regulation of adrenergic receptor responsiveness through modulation of receptor gene expression. Annu. Rev. Physiol. 53: 497-508.
- Dohlman, H.G., et al. 1991. Model systems for the study of seven transmembrane-segment receptors. Annu. Rev. Biochem. 60: 653-688.
- Collins, S., et al. 1992. From ligand binding to gene expression: new insights into the regulation of G protein-coupled receptors. Trends Biochem. Sci. 17: 37-39.
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- Inglese, J., et al. 1994. Functionally active targeting domain of the β-adrenergic receptor kinase: an inhibitor of G_{βy}-mediated stimulation of type II adenylyl cyclase. Proc. Natl. Acad. Sci. USA 91: 3637-3641.

CHROMOSOMAL LOCATION

Genetic locus: ADRB2 (human) mapping to 5g32.

PRODUCT

 $\beta_2\text{-AR}$ 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 $\beta_2\text{-AR}$ shRNA Plasmid (h): sc-39866-SH and $\beta_2\text{-AR}$ shRNA (h) Lentiviral Particles: sc-39866-V as alternate gene silencing products.

For independent verification of β_2 -AR (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-39866A, sc-39866B and sc-39866C.

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

 $\beta_2\text{-AR}$ siRNA (h) is recommended for the inhibition of $\beta_2\text{-AR}$ 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-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

GENE EXPRESSION MONITORING

 β_2 -AR (E-3): sc-271322 is recommended as a control antibody for monitoring of β_2 -AR 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-lgG κ BP-HRP: sc-516102 or m-lgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz MarkerTM Molecular Weight Standards: sc-2035, UltraCruz[®] Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-lgG κ BP-FITC: sc-516140 or m-lgG 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 β_2 -AR gene expression knockdown using RT-PCR Primer: β_2 -AR (h)-PR: sc-39866-PR (20 μ l, 425 bp). 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.