SANTA CRUZ BIOTECHNOLOGY, INC.

A cyclase VIII siRNA (m): sc-40326

BACKGROUND

Adenylyl cyclases function to convert ATP to cyclic AMP in response to activation by a variety of hormones, neurotransmitters and other regulatory molecules. Adenylyl cyclases respond to receptor-initiated signals, mediated by the G_s and G_i heterotrimeric G proteins. The binding of an agonist to a G_s-coupled receptor catalyzes the exchange of GDP (bound to $G_{\alpha s}$) for GTP, dissociation of GTP-$G_{\alpha s}$ from $G_{\beta\gamma}$ and $G_{\alpha s}$-mediated activation of adenylyl cyclase. Adenylyl cyclase type VIII (AC VIII) is one of the three mammalian calcium-stimulated isoforms, each of which is expressed in a region-specific manner in the central nervous system. In addition to the high expression in the brain, A cyclase VIII is also expressed in the lung. Ca^{2+}/calmodulin-dependent A cyclase VIII immunoreactivity is increased in alcoholic corpus amyadaloideum and hippocampus, suggesting that adenyl cyclase may play a role in the pathophysiology of alcoholism. A significant decrease in the level of A cyclase I and a tendency to decrease in the level of A cyclase VIII in Alzheimer's disease hippocampus, suggesting that A cyclase I and VIII may play an essential role in learning and memory. A cyclase VIII knock-out mice do not have normal increases in behavioral markers of anxiety; thus, A cyclase VIII may also function in the modulation of anxiety.

REFERENCES

1. Gilman, A.G. 1987. G proteins: transducers of receptor-generated signals. Annu. Rev. Biochem. 56: 615-649.
2. Bourne, H.R., et al. 1990. The GTPase superfamily: a conserved switch for diverse cell functions. Nature 348: 125-132.
3. Tang, W.J. and Gilman, A.G. 1992. Adenylyl cyclases. Cell 70: 869-872.
4. Taussig, R., et al. 1994. Distinct patterns of bidirectional regulation of mammalian adenylyl cyclases. J. Biol. Chem. 269: 6093-6100.
5. Muglia, L.M., et al. 1999. The 5'-flanking region of the mouse adenylyl cyclase type VIII gene imparts tissue-specific expression in transgenic mice. J. Neurosci. 19: 2051-2058.
6. Yamamoto, M., et al. 2000. Hippocampal level of neural specific adenylyl cyclase type I is decreased in Alzheimer's disease. Biochim. Biophys. Acta 1535: 60-68.

CHROMOSOMAL LOCATION

Genetic locus: Adcy8 (mouse) mapping to 15 D1.

PRODUCT

A cyclase VIII 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 A cyclase VIII shRNA Plasmid (m): sc-40326-SH and A cyclase VIII shRNA (m) Lentiviral Particles: sc-40326-V as alternate gene silencing products.

For independent verification of A cyclase VIII (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-40326A, sc-40326B and sc-40326C.

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

A cyclase VIII siRNA (m) is recommended for the inhibition of A cyclase VIII 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

A cyclase VIII (B-6): sc-377323 is recommended as a control antibody for monitoring of A cyclase VIII 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 A cyclase VIII gene expression knockdown using RT-PCR Primer: A cyclase VIII (m)-PR: sc-40326-PR (20 µl, 420 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.