SANTA CRUZ BIOTECHNOLOGY, INC.

PCP-2 siRNA (m): sc-61308

BACKGROUND

Purkinje cells are densely branching neurons characteristic of the cerebellar cortex. Purkinje cell protein-2 (PCP-2 or L7) is a G protein regulator abundant in Purkinje cells and retinal bipolar neurons. PCP-2 belongs to a family of proteins containing a GoLoco or GPR (G protein regulatory) motif named for the $G_{i/o}$ interacting protein Loco, the *Drosophila* RGS12 homologue. PCP-2 protein interacts with $G_{\alpha\ i/o}$ family of G proteins to inhibit GDP release. This indicates that the colocalization and association of $G_{\alpha\ i/o}$ and PCP-2 in cerebellum may play a functional role in regions of synaptic activity, as well as neural differentiation. The Purkinje type calcium channels may be physiological effectors of PCP-2 because they are the major voltage-dependent channels that modulate cell output and they are regulated by $G_{i/o}$ proteins. PCP-2 is only detected in higher vertebrates, suggesting that it may be a marker of more recent evolutionary development of cerebellar Purkinje cells.

REFERENCES

1. Redd, K.J., et al. 2002. Association and co-localization of G protein α-subunits and Purkinje cell protein-2 (PCP-2) in mammalian cerebellum. J. Neurosci. Res. 70: 631-637.
2. Zhang, X., et al. 2002. Conservation of the developmentally regulated dendritic localization of a Purkinje cell-specific mRNA that encodes a G protein modulator: comparison of rodent and human L7/PCP-2 gene structure and expression. Brain Res. Mol. Brain Res. 105: 1-10.
3. Kinoshita-Kawada, M., et al. 2004. A Purkinje cell-specific GoLoco domain protein, L7/PCP-2, modulates receptor-mediated inhibition of Cav 2.1 Ca^{2+} channels in a dose-dependent manner. Brain Res. Mol. Brain Res. 132: 73-86.
4. Rong, Y., et al. 2004. Identification of candidate Purkinje cell-specific markers by gene expression profiling in wild-type and pcd(3J) mice. Brain Res. Mol. Brain Res. 132: 128-145.
5. Goswami, J., et al. 2005. Enhanced Purkinje cell survival but compromised cerebellar function in targeted anti-apoptotic protein transgenic mice. Mol. Cell. Neurosci. 29: 202-221.
6. Saito, H., et al. 2005. L7/PCP-2-specific expression of Cre recombinase using knock-in approach. Biochem. Biophys. Res. Commun. 331: 1216-1221.

CHROMOSOMAL LOCATION

Genetic locus: Pcp2 (mouse) mapping to 8 A1.1.

PRODUCT

PCP-2 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 PCP-2 shRNA Plasmid (m): sc-61308-SH and PCP-2 shRNA (m) Lentiviral Particles: sc-61308-V as alternate gene silencing products.

For independent verification of PCP-2 (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-61308A and sc-61308B.

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

PCP-2 siRNA (m) is recommended for the inhibition of PCP-2 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

PCP-2 (F-3): sc-137064 is recommended as a control antibody for monitoring of PCP-2 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 PCP-2 gene expression knockdown using RT-PCR Primer: PCP-2 (m)-PR: sc-61308-PR (20 µl). 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.