

P2Y13 siRNA (m): sc-76029

BACKGROUND

Nucleotides are important extracellular signaling molecules that mediate several events, such as cell proliferation, differentiation, chemotaxis and cytokine release. The P2 receptor family is activated by the binding of nucleotides and is divided into two subfamilies, designated P2X and P2Y. The P2Y receptor family are G protein-coupled receptors which mediate the effects of extracellular nucleotides, primarily through the activation of phospholipase C (PLC). To some extent, the P2Y receptors can also activate potassium channels or, alternatively, inhibit adenylate cyclase and N-type calcium channels in response to extracellular nucleotides. P2Y13 (purinergic receptor P2Y, G protein-coupled, 13), also known as GPCR1, GPR86 or GPR94, is a 354 amino acid multi-pass membrane protein that belongs to the P2Y receptor family and exists as two alternatively spliced isoforms. Expressed at high levels in spleen and adult brain tissue, P2Y13 functions as a receptor for ADP and is thought to play a role in immune system activity, as well as in hematopoiesis.

REFERENCES

1. Lee, D.K., Nguyen, T., Lynch, K.R., Cheng, R., Vanti, W.B., Arkhitko, O., Lewis, T., Evans, J.F., George, S.R. and O'Dowd, B.F. 2001. Discovery and mapping of ten novel G protein-coupled receptor genes. *Gene* 275: 83-91.
2. Communi, D., Gonzalez, N.S., Detheux, M., Brezillon, S., Lannoy, V., Parmentier, M. and Boeynaems, J.M. 2001. Identification of a novel human ADP receptor coupled to G_i. *J. Biol. Chem.* 276: 41479-41485.
3. Wittenberger, T., Schaller, H.C. and Hellebrand, S. 2001. An expressed sequence tag (EST) data mining strategy succeeding in the discovery of new G protein-coupled receptors. *J. Mol. Biol.* 307: 799-813.
4. Takeda, S., Kadowaki, S., Haga, T., Takaesu, H. and Mitaku, S. 2002. Identification of G protein-coupled receptor genes from the human genome sequence. *FEBS Lett.* 520: 97-101.
5. Zhang, F.L., Luo, L., Gustafson, E., Palmer, K., Qiao, X., Fan, X., Yang, S., Laz, T.M., Bayne, M. and Monsma, F. 2002. P2Y(13): identification and characterization of a novel G_α_i-coupled ADP receptor from human and mouse. *J. Pharmacol. Exp. Ther.* 301: 705-713.
6. Kim, Y.C., Lee, J.S., Sak, K., Marteau, F., Mamedova, L., Boeynaems, J.M. and Jacobson, K.A. 2005. Synthesis of pyridoxal phosphate derivatives with antagonist activity at the P2Y13 receptor. *Biochem. Pharmacol.* 70: 266-274.

CHROMOSOMAL LOCATION

Genetic locus: P2ry13 (mouse) mapping to 3 D.

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.

PRODUCT

P2Y13 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see P2Y13 shRNA Plasmid (m): sc-76029-SH and P2Y13 shRNA (m) Lentiviral Particles: sc-76029-V as alternate gene silencing products.

For independent verification of P2Y13 (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-76029A, sc-76029B and sc-76029C.

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

P2Y13 siRNA (m) is recommended for the inhibition of P2Y13 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 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.

GENE EXPRESSION MONITORING

P2Y13 (6G12E10): sc-517219 is recommended as a control antibody for monitoring of P2Y13 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).

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor P2Y13 gene expression knockdown using RT-PCR Primer: P2Y13 (m)-PR: sc-76029-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.