

LGR5 siRNA (h): sc-62559

BACKGROUND

G protein-coupled receptors (GPCRs), also designated seven transmembrane (7TM) receptors or heptahelical receptors, interact with G proteins (heterotrimeric GTPases) to synthesize intracellular second messengers, such as diacylglycerol, cyclic AMP, inositol phosphates and calcium ions. Their diverse biological functions range from vision and olfaction to neuronal and endocrine signaling and are involved in many pathological conditions. LGR5 (leucine-rich repeat-containing G protein-coupled receptor 5), also known as GPR49 or GPR67, is a 907 amino acid multi-pass membrane protein that contains 17 leucine-rich repeats and belongs to the G protein-coupled receptor family. Expressed in placenta, skeletal muscle and spinal cord, LGR5 functions as an orphan receptor that is thought to play an important role in embryonic growth control and cellular differentiation. Overexpression of LGR5 is associated with increased tumor susceptibility and malignant transformation, implicating LGR5 as a potent tumor-inducing protein.

REFERENCES

1. McDonald, T., et al. 1998. Identification and cloning of an orphan G protein-coupled receptor of the glycoprotein hormone receptor subfamily. *Biochem. Biophys. Res. Commun.* 247: 266-270.
2. Hsu, S.Y., et al. 1998. Characterization of two LGR genes homologous to gonadotropin and thyrotropin receptors with extracellular leucine-rich repeats and a G protein-coupled, seven-transmembrane region. *Mol. Endocrinol.* 12: 1830-1845.
3. Hsu, S.Y., et al. 2000. The three subfamilies of leucine-rich repeat-containing G protein-coupled receptors (LGR): identification of LGR6 and LGR7 and the signaling mechanism for LGR7. *Mol. Endocrinol.* 14: 1257-1271.
4. Yamamoto, Y., et al. 2003. Overexpression of orphan G protein-coupled receptor, GPR49, in human hepatocellular carcinomas with β -catenin mutations. *Hepatology* 37: 528-533.
5. McClanahan, T., et al. 2006. Identification of overexpression of orphan G protein-coupled receptor GPR49 in human colon and ovarian primary tumors. *Cancer Biol. Ther.* 5: 419-426.
6. Barker, N., 2007. Identification of stem cells in small intestine and colon by marker gene *Lgr5*. *Nature* 449: 1003-1007.

CHROMOSOMAL LOCATION

Genetic locus: LGR5 (human) mapping to 12q21.1.

PRODUCT

LGR5 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see LGR5 shRNA Plasmid (h): sc-62559-SH and LGR5 shRNA (h) Lentiviral Particles: sc-62559-V as alternate gene silencing products.

For independent verification of LGR5 (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-62559A, sc-62559B and sc-62559C.

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

LGR5 siRNA (h) is recommended for the inhibition of LGR5 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-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

GENE EXPRESSION MONITORING

LGR5 (634J2E): sc-517661 is recommended as a control antibody for monitoring of LGR5 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 LGR5 gene expression knockdown using RT-PCR Primer: LGR5 (h)-PR: sc-62559-PR (20 μ l, 395 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Hwang, M., et al. 2019. LGR5 and downstream intracellular signaling proteins play critical roles in the cell proliferation of neuroblastoma, meningioma and pituitary adenoma. *Exp. Neurobiol.* 28: 628-641.

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.