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T1R3 siRNA (m): sc-45325

BACKGROUND

The sense of taste provides animals with valuable information about the quality and nutritional value of food. The four widely accepted categories of taste perception are sweet, bitter, salty and sour. A controversial fifth taste, known as umami or monosodium glutamate (MSG), has also been described. A family of G protein-coupled receptors are involved in taste perception, and includes T1R, which is involved in sweet and umami taste perception, and T2R, which is involved in bitter taste perception. The T1R family consists of three members, T1R1, T1R2 and T1R3. These proteins form heterodimers, which alters the selectivity of the subunits. The T1R2 and T1R3 heterodimer functions as a receptor for sweet taste, and recognizes several sweet-tasting molecules, such as sucrose, saccharin, dulcin and acesulfame-K. The T1R1 and T1R3 heterodimer recognizes L-amino-acids to perceive umami taste. Sweet taste transduction is carried out by two pathways. First, sucrose and other sugars activate Gas via the T1Rs, which activates adenylyl cyclase to generate cAMP. Artificial sweeteners bind to either $G_{\beta\gamma}$ or $G_{\alpha q}$ coupled T1Rs to activate PLC β 2 and generate IP3 and DAG. Both pathways ultimately lead to neurotransmitter release. The mouse T1R3 gene maps to chromosome 4 near the Sac locus, a primary determinant of sweet preference in mice, and it is expressed in a subset of taste cells in circumvallate, foliate and fungiform taste papillae.

REFERENCES

1. Nelson, G., et al. 2001. Mammalian sweet taste receptors. *Cell* 106: 381-390.
2. Montmayeur, J.P., et al. 2001. A candidate taste receptor gene near a sweet taste locus. *Nat. Neurosci.* 4: 492-498.
3. Sainz, E., et al. 2001. Identification of a novel member of the T1R family of putative taste receptors. *J. Neurochem.* 77: 896-903.
4. Margolskee, R.F. 2002. Molecular mechanisms of bitter and sweet taste transduction. *J. Biol. Chem.* 277: 1-4.
5. Li, X., et al. 2002. Human receptors for sweet and umami taste. *Proc. Natl. Acad. Sci. USA* 99: 4692-4696.

CHROMOSOMAL LOCATION

Genetic locus: Tas1r3 (mouse) mapping to 4 E2.

PRODUCT

T1R3 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 T1R3 shRNA Plasmid (m): sc-45325-SH and T1R3 shRNA (m) Lentiviral Particles: sc-45325-V as alternate gene silencing products.

For independent verification of T1R3 (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-45325A, sc-45325B and sc-45325C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

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

T1R3 siRNA (m) is recommended for the inhibition of T1R3 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

T1R3 (G-2): sc-398996 is recommended as a control antibody for monitoring of T1R3 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker™ compatible goat anti-mouse IgG-HRP: sc-2031 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor T1R3 gene expression knockdown using RT-PCR Primer: T1R3 (m)-PR: sc-45325-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

See our web site at www.scbt.com or our catalog for detailed protocols and support products.