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COMTD1 siRNA (m): sc-142492

BACKGROUND

Catechol-O-methyltransferase (COMT) plays a crucial role in the regulation of central dopaminergic systems by catalyzing the inactivation of catecholamines. It is widely distributed in most tissues in soluble and membrane-bound forms. COMT-mediated methylation metabolism of catecholamine neurotransmitters is a first-line detoxification pathway. A Val158Met polymorphism of the COMT gene affects activity of the enzyme and influences performance and efficiency of the prefrontal cortex of the brain. Sequential conversion of estradiol to methoxyestradiol by COMT, contributes to the antimutagenic effects of estradiol on vascular smooth muscle cell growth via estrogen receptor-independent mechanisms. COMTD1 (catechol O-methyltransferase domain-containing protein 1) is a 262 amino acid single-pass type II membrane protein that belongs to the methyltransferase superfamily. A putative O-methyltransferase, COMTD1 exists as a homodimer and is encoded by a gene that maps to human chromosome 10q22.21.

REFERENCES

1. Masuda, M., et al. 2002. Assay of catechol-O-methyltransferase activity in human erythrocytes using norepinephrine as a natural substrate. *Ann. Clin. Biochem.* 39: 589-594.
2. Dubey, R.K., et al. 2004. Catecholamines block the antimutagenic effect of estradiol on human coronary artery smooth muscle cells. *J. Clin. Endocrinol. Metab.* 89: 3922-3931.
3. Tunbridge, E.M., et al. 2004. Catechol-o-methyltransferase inhibition improves set-shifting performance and elevates stimulated dopamine release in the rat prefrontal cortex. *J. Neurosci.* 24: 5331-5335.
4. Zhu, B.T. 2004. CNS dopamine oxidation and catechol-O-methyltransferase: importance in the etiology, pharmacotherapy, and dietary prevention of Parkinson's disease. *Int. J. Mol. Med.* 13: 343-353.
5. Tunbridge, E.M., et al. 2008. Polymorphisms in the catechol-O-methyltransferase (COMT) gene influence plasma total homocysteine levels. *Am. J. Med. Genet. B Neuropsychiatr. Genet.* 147B: 996-999.
6. Hirata, H., et al. 2008. COMT polymorphisms affecting protein expression are risk factors for endometrial cancer. *Mol. Carcinog.* 47: 768-774.

CHROMOSOMAL LOCATION

Genetic locus: Comtd1 (mouse) mapping to 14 A3.

PRODUCT

COMTD1 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 COMTD1 shRNA Plasmid (m): sc-142492-SH and COMTD1 shRNA (m) Lentiviral Particles: sc-142492-V as alternate gene silencing products.

For independent verification of COMTD1 (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-142492A, sc-142492B and sc-142492C.

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

COMTD1 siRNA (m) is recommended for the inhibition of COMTD1 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

COMTD1 (C-5): sc-515490 is recommended as a control antibody for monitoring of COMTD1 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-IgG κ BP-HRP: sc-516102 or m-IgG κ 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-IgG κ BP-FITC: sc-516140 or m-IgG κ 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 COMTD1 gene expression knockdown using RT-PCR Primer: COMTD1 (m)-PR: sc-142492-PR (20 μ l). 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.