β-Gal siRNA (m): sc-61342



The Power to Question

BACKGROUND

The human β -galactosidase gene, known as the LacZ gene, maps to chromosome 3p21.33 and encodes a 677 amino acid protein with an optimum functional pH range of 6 to 8. Catalytically active β -galactosidaseis (β -Gal) is a tetramer of four identical subunits, each with an active site, which can independently catalyze the cleavage of terminal galactose. Monovalent cations have a stimulatory effect on the enzymatic reaction, which likely involves a galactosyl-enzyme complex intermediate. β -Gals are widespread in animals, microorganisms and plants. The LacZ gene is widely used as a reporter gene with a variety of colored or fluorescent compounds capable of being produced from appropriate substrates, such as Xgal, which produces a blue color. For this reason, LacZ is incorporated into numerous plasmid vectors as a marker.

CHROMOSOMAL LOCATION

Genetic locus: Glb1 (mouse) mapping to 9 F3.

PRODUCT

 $\beta\text{-}Gal$ 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 μM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see $\beta\text{-}Gal$ shRNA Plasmid (m): sc-61342-SH and $\beta\text{-}Gal$ shRNA (m) Lentiviral Particles: sc-61342-V as alternate gene silencing products.

For independent verification of β -Gal (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-61342A, sc-61342B and sc-61342C.

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

 $\beta\text{-Gal}$ siRNA (m) is recommended for the inhibition of $\beta\text{-Gal}$ 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

 β -Gal (B-12): sc-377257 is recommended as a control antibody for monitoring of β -Gal 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-lgG κ BP-HRP: sc-516102 or m-lgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz MarkerTM Molecular Weight Standards: sc-2035, UltraCruz[®] Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-lgG κ BP-FITC: sc-516140 or m-lgG 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 β -Gal gene expression knockdown using RT-PCR Primer: β -Gal (m)-PR: sc-61342-PR (20 μ l, 444 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

- Szychowski, K.A., et al. 2019. Impact of elastin-derived VGVAPG peptide on bidirectional interaction between peroxisome proliferator-activated receptor gamma (Pparγ) and β-galactosidase (β-Gal) expression in mouse cortical astrocytes *in vitro*. Naunyn Schmiedebergs Arch. Pharmacol. 392: 405-413.
- Szychowski, K.A., et al. 2019. Impact of elastin-derived peptide VGVAPG on matrix metalloprotease-2 and -9 and the tissue inhibitor of metalloproteinase-1, -2, -3 and -4 mRNA expression in mouse cortical glial cells in vitro. Neurotox. Res. 35: 100-110.
- Szychowski, K.A., et al. 2019. The VGVAPG peptide regulates the production of nitric oxide synthases and reactive oxygen species in mouse astrocyte cells in vitro. Neurochem. Res. 44: 1127-1137.
- 4. Szychowski, K.A., et al. 2019. Elastin-derived peptide VGVAPG affects the proliferation of mouse cortical astrocytes with the involvement of aryl hydrocarbon receptor (Ahr), peroxisome proliferator-activated receptor γ (Ppary), and elastin-binding protein (EBP). Cytokine 126: 154930.
- Szychowski, K.A., et al. 2019. Specific role of N-methyl-D-aspartate (NMDA) receptor in elastin-derived VGVAPG peptide-dependent calcium homeostasis in mouse cortical astrocytes *in vitro*. Sci. Rep. 9: 20165.

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.

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