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Diagnostik & molekulare Diagnostik



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p38α shRNA (m2) Lentiviral Particles: sc-44217-V



The Power to Overtin

BACKGROUND

MAP (mitogen-activated protein) kinases play a significant role in many biological processes, including cell adhesion and spreading, cell differentiation and apoptosis. p38 α , p38 β and p38 γ , also known as MAPK14, MAPK11 and MAPK12, respectively, each contain one protein kinase domain and belong to the MAP kinase family. Expressed in different areas throughout the body with common expression patterns in heart, p38 proteins use magnesium as a cofactor to catalyze the ATP-dependent phosphorylation of target proteins. Via their catalytic activity, p38 α , p38 β and p38 γ are involved in a variety of events throughout the cell, including signal transduction pathways, cytokine production and cell proliferation and differentiation. The p38 proteins are subject to phosphoryation on Thr and Tyr residues, an event which is thought to activate the phosphorylated protein.

REFERENCES

- Lee, J.C., et al. 1994. A protein kinase involved in the regulation of inflammatory cytokine biosynthesis. Nature 372: 739-746.
- 2. Han, J., et al. 1995. Molecular cloning of human p38 MAP kinase. Biochim. Biophys. Acta 1265: 224-227.
- 3. Li, Z., et al. 1996. The primary structure of p38 γ: a new member of p38 group of MAP kinases. Biochem. Biophys. Res. Commun. 228: 334-340.
- Jiang, Y., et al. 1996. Characterization of the structure and function of a new mitogen-activated protein kinase (p38β). J. Biol. Chem. 271: 17920-17926.
- 5. Tamura, K., et al. 2000. Requirement for p38 α in erythropoietin expression: a role for stress kinases in erythropoiesis. Cell 102: 221-231.

CHROMOSOMAL LOCATION

Genetic locus: Mapk14 (mouse) mapping to 17 A3.3.

PRODUCT

p38 α shRNA (m2) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 2 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 μ l frozen stock containing 1.0 x 10 6 infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see p38 α siRNA (m2): sc-44217 and p38 α shRNA Plasmid (m2): sc-44217-SH as alternate gene silencing products.

STORAGE

Store lentiviral particles at -80° C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4° C for up to one week. Avoid repeated freeze thaw cycles.

PROTOCOLS

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

APPLICATIONS

 $p38\alpha$ shRNA (m2) Lentiviral Particles is recommended for the inhibition of p38 expression in mouse cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 µl frozen viral stock containing 1.0 x 10⁶ infectious units of virus (IFU); contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

GENE EXPRESSION MONITORING

 $p38\alpha$ (F-9): sc-271120 is recommended as a control antibody for monitoring of p38 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) Immunofluo-rescence: use goat anti-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor p38 gene expression knockdown using RT-PCR Primer: p38 α (m2)-PR: sc-44217-PR (20 μ I, 422 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.

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