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MuRF1 siRNA (h): sc-43951

BACKGROUND

Muscle specific RING finger protein (MuRF1) is a sarcomere-associated protein that is upregulated by conditions that provoke atrophy. Pharmacological or genetic inhibition of the IKK β /NF κ B/MuRF1 pathway reverses muscle atrophy, which presents MuRF as a target for clinical intervention. MuRF1 is a key regulator of the PKC-dependent hypertrophic response and can blunt cardiomyocyte hypertrophy, which may have important implications in the pathophysiology of clinical cardiac hypertrophy. MuRF1 directly associates with Titin kinase and influences microtubule-dependent signaling pathways in striated muscle and iris. MuRF1 upregulation is an indicator for skeletal muscle atrophy mechanisms that utilize ubiquitin-dependent proteolysis. MuRF1 transcript levels are high in situations where there is an overabundance of reactive oxygen species, such as cancer, AIDS and sepsis.

REFERENCES

- Centner, T., et al. 2001. Identification of muscle specific ring finger proteins as potential regulators of the titin kinase domain. *J. Mol. Biol.* 306: 717-726.
- Bodine, S.C., et al. 2001. Identification of ubiquitin ligases required for skeletal muscle atrophy. *Science* 294: 1704-1708.
- Li, Y.P., et al. 2003. Hydrogen peroxide stimulates ubiquitin-conjugating activity and expression of genes for specific E2 and E3 proteins in skeletal muscle myotubes. *Am. J. Physiol., Cell Physiol.* 285: C806-C812.
- Glass, D.J. 2003. Signalling pathways that mediate skeletal muscle hypertrophy and atrophy. *Nat. Cell Biol.* 5: 87-90.
- Glass, D.J. 2003. Molecular mechanisms modulating muscle mass. *Trends Mol. Med.* 9: 344-350.
- Sachek, J.M., et al. 2004. IGF-I stimulates muscle growth by suppressing protein breakdown and expression of atrophy-related ubiquitin ligases, atrogin-1 and MuRF1. *Am. J. Physiol. Endocrinol. Metab.* 287: E591-E601.

CHROMOSOMAL LOCATION

Genetic locus: TRIM63 (human) mapping to 1p36.11.

PRODUCT

MuRF1 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 MuRF1 shRNA Plasmid (h): sc-43951-SH and MuRF1 shRNA (h) Lentiviral Particles: sc-43951-V as alternate gene silencing products.

For independent verification of MuRF1 (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-43951A, sc-43951B and sc-43951C.

PROTOCOLS

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

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

MuRF1 siRNA (h) is recommended for the inhibition of MuRF1 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

MuRF1 (C-11): sc-398608 is recommended as a control antibody for monitoring of MuRF1 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 MuRF1 gene expression knockdown using RT-PCR Primer: MuRF1 (h)-PR: sc-43951-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.