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- Trockeneiszuschlag
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SANTA CRUZ BIOTECHNOLOGY, INC.

MPO shRNA (m) Lentiviral Particles: sc-43942-V

BACKGROUND

The heme protein myeloperoxidase (MPO) is a major component of azurophilic granules of neutrophils and polymorphonuclear leukocytes. Optimal oxygen-dependent microbiocidal activity depends on MPO as the critical enzyme for the generation of hypochlorous acid and other toxic oxygen products. The MPO precursor is synthesized during the promyelocytic stage of myeloid differentiation and is subsequently processed and transported intracellularly to the lysosomes. The precursor undergoes cotranslational N-linked glycosylation to produce a glycoprotein. Glucosidases in the endoplasmic reticulum (ER) or early cis Golgi convert the pro-MPO to a form which is sorted into a prelysosomal compartment, which undergoes final proteolytic maturation to native MPO, a pair of heavy-light protomers. In normal neutrophils, MPO is expressed as a dimer. Calreticulin, a calcium-binding protein residing in the ER, interacts specifically with fully glycosylated apopro-MPO. iMPO mRNA is abundant in human promyelocytic HL-60 and mouse myeloid leukemia NFS-60 cells. MPO is expressed at high levels in circulating neutrophils and monocytes but is not detectable in microglia, brain-specific macrophages or normal brain tissue.

REFERENCES

1. Johnson, K.R., et al. 1987. Characterization of cDNA clones for human myeloperoxidase: predicted amino acid sequence and evidence for multiple mRNA species. Nucleic Acids Res. 15: 2013-2028.
2. Nauseef, W.M. 1987. Postranslational processing of a human myeloid lysosomal protein, myeloperoxidase. Blood 70: 1143-1150.
3. Morishita, K., et al. 1987. Molecular cloning and characterization of cDNA for human myeloperoxidase. J. Biol. Chem. 262: 3844-3851.
4. Nauseef, W.M., et al. 1988. Biosynthesis and processing of myeloperoxidase a marker for myeloid cell differentiation. Eur. J. Haematol. 40: 97-110.

CHROMOSOMAL LOCATION

Genetic locus: Mpo (mouse) mapping to 11 C.

PRODUCT

MPO shRNA (m) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 3 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 µl 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 MPO siRNA (m): sc-43942 and MPO shRNA Plasmid (m): sc-43942-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

MPO shRNA (m) Lentiviral Particles is recommended for the inhibition of MPO expression in mouse cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 µl frozen viral stock containing 1.0 x 10^6 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

MPO light chain (C-3): sc-390109 is recommended as a control antibody for monitoring of MPO 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 MPO gene expression knockdown using RT-PCR Primer: MPO (m)-PR: sc-43942-PR (20 µl). 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.