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Iba1 shRNA (h) Lentiviral Particles: sc-43857-V

BACKGROUND

Ionized calcium-binding adapter molecule 1 (Iba1), also known as allograft inflammatory factor-1 (AIF-1), is a 147 amino acid cytoplasmic, calcium-binding protein that is thought to play a role in macrophage activation and function. Iba1, containing two EF domains, is induced by cytokines and interferons. In an unstimulated state, Iba1 colocalizes with actin, and upon stimulation, translocates to lamellipodia. It is also a marker of human microglia and is expressed by macrophages in injured skeletal muscle. The gene encoding Iba1 maps to chromosome 6p21.33 and resides in the tumor necrosis factor (TNF) cluster of genes located in the region represented by the human major histocompatibility complex (MHC).

REFERENCES

1. Autieri, M.V. 1996. cDNA cloning of human allograft inflammatory factor-1: tissue distribution, cytokine induction, and mRNA expression in injured rat carotid arteries. *Biochem. Biophys. Res. Commun.* 228: 29-37.
2. Autieri, M.V. and Agrawal, N. 1998. IRT-1, a novel interferon- γ -responsive transcript encoding a growth-suppressing basic leucine zipper protein. *J. Biol. Chem.* 273: 14731-14737.
3. Neville, M.J. and Campbell, R.D. 1999. A new member of the Ig superfamily and a V-ATPase G subunit are among the predicted products of novel genes close to the TNF locus in the human MHC. *J. Immunol.* 162: 4745-4754.
4. Autieri, M.V., Kelemen, S.E. and Wendt, K.W. 2003. AIF-1 is an actin-polymerizing and Rac1-activating protein that promotes vascular smooth muscle cell migration. *Circ. Res.* 92: 1107-1114.

CHROMOSOMAL LOCATION

Genetic locus: AIF1 (human) mapping to 6p21.33.

PRODUCT

Iba1 shRNA (h) Lentiviral Particles are concentrated, transduction-ready viral particles containing a target-specific construct that encodes a 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 μ l frozen stock containing 1.0×10^6 lentiviral transducing particles per milliliter in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see Iba1 siRNA (h): sc-43857 and Iba1 shRNA Plasmid (h): sc-43857-SH as alternate gene silencing products.

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.

APPLICATIONS

Iba1 shRNA (h) Lentiviral Particles is recommended for the inhibition of Iba1 expression in human cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μ l frozen viral stock containing 1.0×10^6 lentiviral transducing particles per milliliter; contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

GENE EXPRESSION MONITORING

Iba1 (F-4): sc-398406 is recommended as a control antibody for monitoring of Iba1 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 Iba1 gene expression knockdown using RT-PCR Primer: Iba1 (h)-PR: sc-43857-PR (20 μ l). 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.

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.