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β-2-Microglobulin shRNA (h2) Lentiviral Particles: sc-44280-V

BACKGROUND

Major histocompatibility complex (MHC) class 1 molecules bind to antigens for presentation on the surface of cells. The proteasome is responsible for producing these antigens from the components of foreign pathogens. MHC class 1 molecules consist of an α heavy chain that contains three subdomains ($\alpha 1$, $\alpha 2$, $\alpha 3$), and a non-covalent associating light chain, known as β -2-Microglobulin. β -2-Microglobulin associates with the $\alpha 3$ subdomain of the α heavy chain and forms an immunoglobulin domain-like structure that mediates proper folding and expression of MHC class 1 molecules. The $\alpha 1$ and $\alpha 2$ domains of the α heavy chain form the peptide antigen-binding cleft. Mice that lack β -2-Microglobulin protein show a normal distribution of T cells, yet have no mature CD4-8⁺ T cells and are defective in CD4-8⁺ T cell-mediated cytotoxicity. Interferon- γ can stimulate production of β -2-Microglobulin transcripts. The human β -2-Microglobulin gene maps to chromosome 15q21-q22.2 and encodes a 119 amino acid protein. Mutations in the β -2-Microglobulin gene can enhance the progression of malignant melanoma phenotypes.

REFERENCES

1. Skjødtt, K., et al. 1987. Isolation and characterization of chicken and turkey β -2-Microglobulin. *Mol. Immunol.* 23: 1301-1309.
2. Dunon, D., et al. 1990. T cell precursor migration towards β -2-Microglobulin is involved in thymus colonization of chicken embryos. *EMBO J.* 9: 3315-3322.
3. Zijlstra, M., et al. 1990. β 2-Microglobulin deficient mice lack CD4-8⁺ cytolytic T cells. *Nature* 344: 742-746.

CHROMOSOMAL LOCATION

Genetic locus: B2M (human) mapping to 15q21.1.

PRODUCT

β -2-Microglobulin shRNA (h2) 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 μ l frozen stock containing 1.0×10^6 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 β -2-Microglobulin siRNA (h2): sc-44280 and β -2-Microglobulin shRNA Plasmid (h2): sc-44280-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

β -2-Microglobulin shRNA (h2) Lentiviral Particles is recommended for the inhibition of β -2-Microglobulin 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 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

β -2-Microglobulin (BBM.1): sc-13565 is recommended as a control antibody for monitoring of β -2-Microglobulin 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 β -2-Microglobulin gene expression knockdown using RT-PCR Primer: β -2-Microglobulin (h2)-PR: sc-44280-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.