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CD164 shRNA (m) Lentiviral Particles: sc-44678-V

BACKGROUND

CD164 is a mucin-like cell surface glycoprotein that facilitates adhesion of CD34⁺ cells and serves as a negative regulator of hematopoietic progenitor cell proliferation. Human CD164 in CD34⁺CD38⁺ hematopoietic progenitor and epithelial cell lines localizes to endosomes and lysosomes, with low concentrations also appearing at the cell surface.

REFERENCES

1. Watt, S.M., et al. 1998. CD164, a novel sialomucin on CD34⁺ and erythroid subsets, is located on human chromosome 6q21. *Blood* 92: 849-866.
2. Doyonnas, R., et al. 2000. CD164 monoclonal antibodies that block hematopoietic progenitor cell adhesion and proliferation interact with the first mucin domain of the CD164 receptor. *J. Immunol.* 165: 840-851.
3. Watt, S.M., et al. 2000. Functionally defined CD164 epitopes are expressed on CD34⁺ cells throughout ontogeny but display distinct distribution patterns in adult hematopoietic and nonhematopoietic tissues. *Blood* 95: 3113-3124.
4. Chan, J.Y., et al. 2001. Relationship between novel isoforms, functionally important domains, and subcellular distribution of CD164/endolyn. *J. Biol. Chem.* 276: 2139-2152.
5. Lee, Y.N., et al. 2001. Identification of a role for the sialomucin CD164 in myogenic differentiation by signal sequence trapping in yeast. *Mol. Cell. Biol.* 21: 7696-7706.
6. Jorgensen-Tye, B., et al. 2005. Epitope recognition of antibodies that define the sialomucin, endolyn (CD164), a negative regulator of haematopoiesis. *Tissue Antigens* 65: 220-239.

CHROMOSOMAL LOCATION

Genetic locus: Cd164 (mouse) mapping to 10 B2.

PRODUCT

CD164 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 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 CD164 siRNA (m): sc-44678 and CD164 shRNA Plasmid (m): sc-44678-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

CD164 shRNA (m) Lentiviral Particles is recommended for the inhibition of CD164 expression in mouse 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

CD164 (M-197): sc-33125 is recommended as a control antibody for monitoring of CD164 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-rabbit IgG-HRP: sc-2004 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible goat anti-rabbit IgG-HRP: sc-2030 (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-rabbit IgG-FITC: sc-2012 (dilution range: 1:100-1:400) or goat anti-rabbit IgG-TR: sc-2780 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor CD164 gene expression knockdown using RT-PCR Primer: CD164 (m)-PR: sc-44678-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.

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.