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# Clusterin shRNA (h) Lentiviral Particles: sc-43688-V

## BACKGROUND

Clusterin, also designated complement lysis inhibitor (CLI), apolipoprotein J (ApoJ), sulfated glycoprotein 2 (SGP2), SP-40 and testosterone-repressed prostate message 2 (TRPM2), is a secretory, heterodimeric glycoprotein that influences immune regulation, cell adhesion, transformation, lipid transportation, tissue remodeling, membrane recycling and cell-cell interactions. Clusterin is synthesized as a 449 amino acid polypeptide that is post-translationally cleaved at an internal bond between Arg 227 and Ser 228. Two subunits,  $\alpha$  and  $\beta$ , are associated through disulfide bonds. The  $\beta$  subunit (also called ApoJ $\alpha$ ) corresponds to residues 23-227. The  $\alpha$  subunit (also called ApoJ $\beta$ ) corresponds to residues 228-449. Overexpression of Clusterin appears to be more common in late stages of mammary tumor progression. Clusterin markedly influences  $\beta$ -Amyloid structure and neuritic toxicity *in vivo* and may influence Alzheimer's disease pathogenesis.

## REFERENCES

- de Silva, H.V., et al. 1990. Apolipoprotein J: structure and tissue distribution. *Biochemistry* 29: 5380-5389.
- Rosenberg, M.E., et al. 2002. Apolipoprotein J/clusterin prevents a progressive glomerulopathy of aging. *Mol. Cell. Biol.* 22: 1893-1902.
- Chen, X., et al. 2003. Clusterin as a biomarker in murine and human intestinal neoplasia. *Proc. Natl. Acad. Sci. USA* 100: 9530-9535.
- Leskov, K.S., et al. 2003. Synthesis and functional analyses of nuclear Clusterin, a cell death protein. *J. Biol. Chem.* 278: 11590-11600.
- Gwon, J.S., et al. 2004. Expression of Clusterin in Muller cells of the rat retina after pressure-induced ischemia. *Glia* 47: 35-45.

## CHROMOSOMAL LOCATION

Genetic locus: CLU (human) mapping to 8p21.1.

## PRODUCT

Clusterin shRNA (h) 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  $\mu$ l frozen stock containing  $1.0 \times 10^6$  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 Clusterin siRNA (h): sc-43688 and Clusterin shRNA Plasmid (h): sc-43688-SH as alternate gene silencing products.

## STORAGE

Store lentiviral particles at  $-80^\circ\text{C}$ . Stable for at least one year from the date of shipment. Once thawed, particles can be stored at  $4^\circ\text{C}$  for up to one week. Avoid repeated freeze thaw cycles.

## PROTOCOLS

See our web site at [www.scbt.com](http://www.scbt.com) or our catalog for detailed protocols and support products.

## APPLICATIONS

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

## SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200  $\mu$ l frozen viral stock containing  $1.0 \times 10^6$  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

Clusterin (A-9): sc-166907 is recommended as a control antibody for monitoring of Clusterin 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<sup>™</sup> compatible goat anti-mouse IgG-HRP: sc-2031 (dilution range: 1:2000-1:5000), Cruz Marker<sup>™</sup> 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<sup>™</sup> Mounting Medium: sc-24941.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Clusterin gene expression knockdown using RT-PCR Primer: Clusterin (h)-PR: sc-43688-PR (20  $\mu$ l, 407 bp). Annealing temperature for the primers should be  $55-60^\circ\text{C}$  and the extension temperature should be  $68-72^\circ\text{C}$ .

## 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.