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Cbl shRNA (h2) Lentiviral Particles: sc-44254-V

BACKGROUND

Cbl (also designated the c-Cbl proto-oncogene, E3 ubiquitin-protein ligase CBL, Casitas B-lineage lymphoma proto-oncogene and RING finger protein 55) has been identified as the cellular homolog of the v-Cbl oncogene isolated from an NFS/N mouse that developed a pre-B cell lymphoma following infection with the replication-competent Cas Br-M murine leukemic virus. c-Cbl is expressed at relatively high levels in a wide range of hematopoietic tumor cell lines as well as in normal tissues such as thymus and testis. The c-Cbl gene product has been identified as a cytoplasmic protein with apparent DNA binding and dimerization domains characteristic of transcription factors. A single c-Cbl locus termed CBL2 has been mapped to human chromosome 11q23. This region of chromosome 11 is involved in translocations and deletions in a broad range of leukemias; c-Cbl has been found to be translocated from chromosome 11 in leukemias with either t(4;11) or t(11;14) abnormalities. Two proteins related to c-Cbl have been identified as Cbl-b (RING finger protein 56) and Cbl-3 (RING finger protein 57). Cbl-b has a proline-rich domain, a nuclear localization signal, a C3HC4 zinc finger and a putative leucine zipper. Cbl-b is expressed in normal and malignant mammary epithelial cells, various normal tissues and hematopoietic tissue and cell lines. Data suggests that Cbl-b encodes a protein that can interact with signal transduction proteins to regulate their function or be regulated by them.

REFERENCES

1. Yunis, J.J., et al. 1986. Prognostic significance of chromosomal abnormalities in acute leukaemias and myelodysplastic syndromes. *Clin. Haematol.* 15: 597-620.
2. Langdon, W.Y., et al. 1989. v-Cbl, an oncogene from a dual-recombinant murine retrovirus that induces early B-lineage lymphomas. *Proc. Natl. Acad. Sci. USA* 86: 1168-1172.
3. Langdon, W.Y., et al. 1989. The c-Cbl proto-oncogene is preferentially expressed in thymus and testis tissue and encodes a nuclear protein. *J. Virol.* 63: 5420-5424.

CHROMOSOMAL LOCATION

Genetic locus: CBL (human) mapping to 11q23.3.

PRODUCT

Cbl 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 Cbl siRNA (h2): sc-44254 and Cbl shRNA Plasmid (h2): sc-44254-SH as alternate gene silencing products.

APPLICATIONS

Cbl shRNA (h2) Lentiviral Particles is recommended for the inhibition of Cbl 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

Cbl (C-3): sc-393543 is recommended as a control antibody for monitoring of 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 Cbl gene expression knockdown using RT-PCR Primer: Cbl (h2)-PR: sc-44254-PR (20 μ l, 408 bp). 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.

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.