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# Sp1 shRNA (h2) Lentiviral Particles: sc-44221-V

## BACKGROUND

Sp1 is a sequence-specific transcription factor that recognizes GGGGCGGGGC and closely related sequences, which are often referred to as GC boxes. Sp1 was initially identified as a HeLa cell-derived factor that selectively activates *in vitro* transcription from the SV40 promoter and binds to the multiple GC boxes in the 21-bp repeated elements in SV40. The sequence specificity of DNA binding is conferred by Zn (II) fingers, whereas a different region of Sp1 appears to regulate the affinity of DNA binding. Sp1 belongs to a subgroup of transcription factors that are phosphorylated upon binding to promoter sequences. Evidence suggests that the early growth response gene, Erg-1 (also known as Zif268 or NGF1-A), may downregulate certain mammalian gene promoters by competing with Sp1 for binding to an overlapping binding motif. The gene encoding human Sp1 maps to chromosome 12q13.13.

## REFERENCES

1. Dynan, W.S., et al. 1983. Isolation of transcription factors that discriminate between different promoters recognized by RNA polymerase II. *Cell* 32: 669-680.
2. Dynan, W.S., et al. 1983. The promoter-specific transcription factor Sp1 binds to upstream sequences in the SV40 early promoter. *Cell* 35: 79-87.
3. Kadonaga, J.T., et al. 1987. Isolation of cDNA encoding transcription factor Sp1 and functional analysis of the DNA binding domain. *Cell* 51: 1079-1090.
4. Kadonaga, J.T., et al. 1988. Promoter-selective activation of transcription by Spl. In Franza, B.R. Jr., Cullen, B.R. and Wong-Staal, F., eds., *The Control of Human Retrovirus Gene Expression*. Cold Spring Harbor, NY: Cold Spring Harbor Laboratory, 239-250.
5. Kadonaga, J.T., et al. 1988. Distinct regions of Sp1 modulate DNA binding and transcriptional activation. *Science* 242: 1566.
6. Jackson, S.P., et al. 1990. GC box binding induces phosphorylation of Sp1 by a DNA-dependent protein kinase. *Cell* 63: 155-165.

## CHROMOSOMAL LOCATION

Genetic locus: SP1 (human) mapping to 12q13.13.

## PRODUCT

Sp1 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  $\mu$ 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 Sp1 siRNA (h2): sc-44221 and Sp1 shRNA Plasmid (h2): sc-44221-SH as alternate gene silencing products.

## STORAGE

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

## APPLICATIONS

Sp1 shRNA (h2) Lentiviral Particles is recommended for the inhibition of Sp1 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

Sp1 (E-3): sc-17824 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 Sp1 gene expression knockdown using RT-PCR Primer: Sp1 (h2)-PR: sc-44221-PR (20  $\mu$ l, 458 bp). Annealing temperature for the primers should be  $55-60^{\circ}$  C and the extension temperature should be  $68-72^{\circ}$  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.

## PROTOCOLS

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