SANTA CRUZ BIOTECHNOLOGY, INC.

# Ly-6D siRNA (h): sc-77553

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*The Power to Question*

## BACKGROUND

Ly-6D (lymphocyte antigen 6D), also known as E48 antigen, is a 128 amino acid glycoprotein that is expressed in squamous cell carcinoma cell lines and squamous cell epithelia tissue. Ly-6D contains a signal peptide, two theoretical phosphorylation sites and three putative myristoylation sites. Upregulation of the gene encoding Ly-6D in head and neck cancers is associated with poor prognosis and high expression of Ly-6D has been linked to enhanced cell migration. Ly-6D is frequently used as a molecular marker for diagnosis and therapy of head-and-neck squamous cell carcinoma (HNSCC). It has been suggested that Ly-6D may regulate the expression levels of certain fucosylated E-selectin ligands and protein FX, a protein that contributes to the last step in the synthesis of GDP-L-fucose, in HNSCC cell lines. This finding is indicative that Ly-6D may regulate tumor cell adhesion in inflamed vessel walls that express E-selectin.

## REFERENCES


1. Brakenhoff, R.H., et al. 1995. The human E48 antigen, highly homologous to the murine Ly-6 antigen ThB, is a GPI-anchored molecule apparently involved in keratinocyte cell-cell adhesion. J. Cell Biol. 129: 1677-1689.
2. Brakenhoff, R.H., et al. 1997. A gain of novel tissue specificity in the human Ly-6 gene E48. J. Immunol. 159: 4879-4886.
3. Shan, X., et al. 1998. Characterization and mapping to human chromosome 8q24.3 of Ly-6-related gene 9804 encoding an apparent homologue of mouse TSA-1. J. Immunol. 160: 197-208.
4. Eshel, R., et al. 2000. The GPI-linked Ly-6 antigen E48 regulates expression levels of the FX enzyme and of E-selectin ligands on head and neck squamous carcinoma cells. J. Biol. Chem. 275: 12833-12840.
5. Online Mendelian Inheritance in Man, OMIM™. 2001. Johns Hopkins University, Baltimore, MD. MIM Number: 606204. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/
6. Tsukada, Y., et al. 2002. Expression of Ly-6D on the surface of normal and neoplastic mammary epithelial cells of the mouse. Jpn. J. Cancer Res. 93: 986-993.


## CHROMOSOMAL LOCATION

Genetic locus: LY6D (human) mapping to 8q24.3.

## PRODUCT

Ly-6D siRNA (h) is a pool of 3 target-specific 19-25 nt siRNAs designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Ly-6D shRNA Plasmid (h): sc-77553-SH and Ly-6D shRNA (h) Lentiviral Particles: sc-77553-V as alternate gene silencing products.

For independent verification of Ly-6D (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-77553A, sc-77553B and sc-77553C.

## STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20° C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20° C, avoid contact with RNAses and repeated freeze thaw cycles.

Resuspend lyophilized siRNA duplex in 330 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

## APPLICATIONS

Ly-6D siRNA (h) is recommended for the inhibition of Ly-6D expression in human cells.

## SUPPORT REAGENTS

For optimal siRNA transfection efficiency, Santa Cruz Biotechnology's siRNA Transfection Reagent: sc-29528 (0.3 ml), siRNA Transfection Medium: sc-36868 (20 ml) and siRNA Dilution Buffer: sc-29527 (1.5 ml) are recommended. Control siRNAs or Fluorescein Conjugated Control siRNAs are available as 10 µM in 66 µl. Each contain a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA. Fluorescein Conjugated Control siRNAs include: sc-36869, sc-44239, sc-44240 and sc-44241. Control siRNAs include: sc-37007, sc-44230, sc-44231, sc-44232, sc-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

## GENE EXPRESSION MONITORING

Ly-6D (C-8): sc-373838 is recommended as a control antibody for monitoring of Ly-6D 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 reagents are recommended: 1) Western Blotting: use m-IgGκ BP-HRP: sc-516102 or m-IgGκ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™ Molecular Weight Standards: sc-2035, UltraCruz® Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgGκ BP-FITC: sc-516140 or m-IgGκ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Ly-6D gene expression knockdown using RT-PCR Primer: Ly-6D (h)-PR: sc-77553-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

## RESEARCH USE

For research use only, not for use in diagnostic procedures.

## PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.