SANTA CRUZ BIOTECHNOLOGY, INC.

Bcl-$x_{S/L}$ siRNA (h2): sc-44246

SANTA CRUZ BIOTECHNOLOGY

The Power to Question

BACKGROUND

The Bcl-2 gene was isolated at the chromosomal breakpoint of t(14;18) bearing follicular B cell lymphomas. Bcl-2 blocks cell death following a variety of stimuli and confers a death-sparing effect to certain hematopoietic cell lines following growth factor withdrawal. A second protein, designated Bcl-associated X protein (Bax) p21, has extensive amino acid homology with Bcl-2 and both homodimerizes and heterodimerizes with Bcl-2. Over-expression of Bax accelerates apoptotic death induced by cytokine deprivation in an IL-3-dependent cell line, and Bax also counters the death repressor activity of Bcl-2. Bcl-x, one of several additional proteins with sequence homology to Bcl-2, is expressed as Bcl-x_L, a 233 amino acid protein with 43% sequence identity with Bcl-2 that suppresses cell death, and Bcl-x_S, a shorter variant that is 178 amino acids in length and lacks a 63 amino acid region (amino acids 126-188) found in Bcl-x_L and which functions as a dominant inhibitor of Bcl-2. A further apoptosis-inducing protein, Bad, dimerizes both with Bcl-x_L and to a lesser extent with Bcl-2, thus displacing Bax and inducing apoptosis.

REFERENCES

1. Nunez, G., et al. 1990. Deregulated Bcl-2 gene expression selectively prolongs survival of growth factor-deprived hemopoietic cell lines. J. Immunol. 144: 3602-3610.
2. Hockenbery, D.M., et al. 1991. Bcl-2 protein is topographically restricted in tissues characterized by apoptotic cell death. Proc. Natl. Acad. Sci. USA 88: 6961-6965.
3. Boise, L.H., et al. 1993. Bcl-x, a Bcl-2-related gene that functions as a dominant regulator of apoptotic cell death. Cell 74: 597-608.
4. Oltvai, Z.N., et al. 1993. Bcl-2 heterodimerizes *in vivo* with a conserved homolog, Bax, that accelerates programmed cell death. Cell 74: 609-619.
5. Gottschalk, A.R., et al. 1994. Identification of immunosuppressant-induced apoptosis in a murine B-cell line and its prevention by Bcl-x but not Bcl-2. Proc. Natl. Acad. Sci. USA 91: 7350-7354.
6. Sato, T., et al. 1994. Interactions among members of the Bcl-2 protein family analyzed with a yeast two-hybrid system. Proc. Natl. Acad. Sci. USA 91: 9238-9242.

CHROMOSOMAL LOCATION

Genetic locus: BCL2L1 (human) mapping to 20q11.21.

PRODUCT

Bcl-$x_{S/L}$ siRNA (h2) 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 Bcl-$x_{S/L}$ shRNA Plasmid (h2): sc-44246-SH and Bcl-$x_{S/L}$ shRNA (h2) Lentiviral Particles: sc-44246-V as alternate gene silencing products.

For independent verification of Bcl-$x_{S/L}$ (h2) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-44246A, sc-44246B and sc-44246C.

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

Bcl-$x_{S/L}$ siRNA (h2) is recommended for the inhibition of Bcl-$x_{S/L}$ 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

Bcl-x_L (H-5): sc-8392 is recommended as a control antibody for monitoring of Bcl-$x_{S/L}$ 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 Bcl-$x_{S/L}$ gene expression knockdown using RT-PCR Primer: Bcl-$x_{S/L}$ (h2)-PR: sc-44246-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.