

# Pbx 4 siRNA (h): sc-38802

## BACKGROUND

Pbx 1, 2, 3 and 4 are members of the TALE (three amino acid loop extension) family of homeodomain-containing proteins. Human pre-B cell acute leukemias are frequently associated with a t(1;19)(q23;p13.3) chromosomal rearrangement which creates a chimeric gene encoding a fusion between the E2A and Pbx 1 gene products. Pbx 2 and Pbx 3 share 92% and 94% respective identities with Pbx 1 over a 266 amino acid region flanking their homeobox domains, while all three proteins are quite divergent at their amino and carboxy termini. Two forms of Pbx 1 and Pbx 3 each differ primarily in their carboxy termini and result from alternative mRNA splicings. Unlike other homeotic selector genes which are expressed transiently during development and differentiation, Pbx gene transcripts are ubiquitously expressed in both fetal and adult tissues and cell lines. Additionally, Pbx 2 and Pbx 3 transcripts are detected in lymphoid cells, which do not express Pbx 1. Pbx 4 expressions is confined to the testis, especially to spermatocytes in the pachytene stage of the first meiotic prophase.

## REFERENCES

1. Nourse, J., et al. 1990. Chromosomal translocation t(1;19) results in synthesis of a homeobox fusion mRNA that codes for a potential chimeric transcription factor. *Cell* 60: 535-545.
2. Kamps, M.P., et al. 1990. A new homeobox gene contributes the DNA binding domain of the t(1;19) translocation protein in pre-B ALL. *Cell* 60: 547-555.
3. Monica, K., et al. 1991. Pbx 2 and Pbx 3, new homeobox genes with extensive homology to the human proto-oncogene Pbx 1. *Mol. Cell. Biol.* 11: 6149-6157.
4. LeBrun, D.P., et al. 1994. Fusion with E2A alters the transcriptional properties of the homeodomain protein Pbx 1 in t(1;19) leukemias. *Oncogene* 9: 1641-1647.
5. Lu, Q., et al. 1994. Fusion with E2A converts the Pbx 1 homeodomain protein into a constitutive transcriptional activator in human leukemias carrying the t(1;19) translocation. *Mol. Cell. Biol.* 14: 3938-3948.
6. Monica, K., et al. 1994. Transformation properties of the E2A-Pbx 1 chimeric oncoprotein: fusion with E2A is essential, but the Pbx 1 homeodomain is dispensable. *Mol. Cell. Biol.* 14: 8304-8314.

## CHROMOSOMAL LOCATION

Genetic locus: PBX4 (human) mapping to 19p13.11.

## PRODUCT

Pbx 4 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Pbx 4 shRNA Plasmid (h): sc-38802-SH and Pbx 4 shRNA (h) Lentiviral Particles: sc-38802-V as alternate gene silencing products.

For independent verification of Pbx 4 (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-38802A, sc-38802B and sc-38802C.

## 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  $\mu$ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330  $\mu$ l of RNase-free water makes a 10  $\mu$ M solution in a 10  $\mu$ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

## APPLICATIONS

Pbx 4 siRNA (h) is recommended for the inhibition of Pbx 4 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  $\mu$ M in 66  $\mu$ l. 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

Pbx 1/2/3/4 (F-3): sc-28313 is recommended as a control antibody for monitoring of Pbx 4 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-IgG $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  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-IgG $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  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 Pbx 4 gene expression knockdown using RT-PCR Primer: Pbx 4 (h)-PR: sc-38802-PR (20  $\mu$ l). 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](http://www.scbt.com) for detailed protocols and support products.