SANTA CRUZ BIOTECHNOLOGY, INC.

Barhl1 siRNA (h): sc-62008

BACKGROUND

Drosophila gene BarH1 contains a homeobox required for external sensory organ fate determination. Homeobox proteins are regulators of place-dependent morphogenesis and play important roles in controlling the expression patterns of cell adhesion molecules. Barhl1 (BarH-like-1) is hypothesized to play a similar role in mouse and human development. *In situ* hybridization of mouse tissues at various stages of development demonstrate that Barhl1 expression is limited to restricted domains of the developing central nervous system, in particular the diencephalon and rhombencephalon. In the developing CNS, the expression of Barhl1 in migrating cells gives rise to the cerebellar external granular layer.

REFERENCES

1. Bulfone, A., et al. 2000. Barhl1, a gene belonging to a new subfamily of mammalian homeobox genes, is expressed in migrating neurons of the CNS. Hum. Mol. Genet. 9: 1443-1452.
2. Blair, I.P., et al. 2002. Search for genes involved in Joubert syndrome: evidence that one or more major loci are yet to be identified and exclusion of candidate genes EN1, EN2, FGF8, and BARHL1. Am. J. Med. Genet. 107: 190-196.
3. Li, S., et al. 2004. Barhl1 regulates migration and survival of cerebellar granule cells by controlling expression of the neurotrophin-3 gene. J. Neurosci. 24: 3104-3114.
4. Gould, D.B., et al. 2004. Mutational analysis of BARHL1 and BARX1 in three new patients with Joubert syndrome. Am. J. Med. Genet. A 131: 205-208.
5. Sud, R., et al. 2005. Transcriptional regulation by Barhl1 and Brn-3c in organ-of-Corti-derived cell lines. Brain Res. Mol. Brain Res. 141: 174-180.
6. Offner, N., et al. 2005. The pro-apoptotic activity of a vertebrate Bar-like homeobox gene plays a key role in patterning the *Xenopus* neural plate by limiting the number of chordin- and shh-expressing cells. Development 132: 1807-1818.
7. Colombo, A., et al. 2006. Zebrafish BarH-like genes define discrete neural domains in the early embryo. Gene Expr. Patterns 6: 347-352.

CHROMOSOMAL LOCATION

Genetic locus: BARHL1 (human) mapping to 9q34.13.

PRODUCT

Barhl1 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 Barhl1 shRNA Plasmid (h): sc-62008-SH and Barhl1 shRNA (h) Lentiviral Particles: sc-62008-V as alternate gene silencing products.

For independent verification of Barhl1 (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-62008A, sc-62008B and sc-62008C.

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

Barhl1 siRNA (h) is recommended for the inhibition of Barhl1 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

Barhl1 (KA.25): sc-130465 is recommended as a control antibody for monitoring of Barhl1 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 Barhl1 gene expression knockdown using RT-PCR Primer: Barhl1 (h)-PR: sc-62008-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.