

PREP-2 siRNA (h): sc-62856

BACKGROUND

PREP-2 (Pbx-regulating protein-2), also known as PBX/knotted 1 homeobox 2 or PKNOX2, is a widely expressed protein belonging to the TALE (three amino acid loop extension)/MEIS family. PREP-2 is a DNA-binding protein that forms stable complexes with Pbx proteins. It is highly homologous to the related protein PREP-1, but displays a more restricted tissue distribution and a higher DNA-dissociation rate. Like PREP-1, PREP-2 forms a heterodimer with Pbx 1. The PREP-2-Pbx 1 dimer is relocated to the nucleus where it associates with HoxB1 to form a ternary complex. In contrast with PREP-1, which acts to increase transcriptional activation in this ternary complex, PREP-2 leads to a slight decrease in transcriptional activity of the ternary complex. Multiple isoforms exist for PREP-2, localizing to the nucleus or cytoplasm. Cytoplasmic isoforms are believed to colocalize with F-Actin, G-Actin and tubulin/microtubules.

REFERENCES

1. Imoto, I., et al. 2001. Identification and characterization of human PKNOX2, a novel homeobox-containing gene. *Biochem. Biophys. Res. Commun.* 287: 270-276.
2. Haller, K., et al. 2002. Prep2: cloning and expression of a new prep family member. *Dev. Dyn.* 225: 358-364.
3. Fognani, C., et al. 2002. Characterization of PREP2, a paralog of PREP, which defines a novel sub-family of the MEINOX TALE homeodomain transcription factors. *Nucleic Acids Res.* 30: 2043-2051.
4. Haller, K., et al. 2004. Subcellular localization of multiple PREP2 isoforms is regulated by Actin, Tubulin, and nuclear export. *J. Biol. Chem.* 279: 49384-49394.
5. Villaescusa, J.C., et al. 2004. Expression of Hox cofactor genes during mouse ovarian follicular development and oocyte maturation. *Gene* 330: 1-7.
6. Mee, L., et al. 2005. Hydrolethalus syndrome is caused by a missense mutation in a novel gene HYL1. *Hum. Mol. Genet.* 14: 1475-1488.

CHROMOSOMAL LOCATION

Genetic locus: PKNOX2 (human) mapping to 11q24.2.

PRODUCT

PREP-2 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see PREP-2 shRNA Plasmid (h): sc-62856-SH and PREP-2 shRNA (h) Lentiviral Particles: sc-62856-V as alternate gene silencing products.

For independent verification of PREP-2 (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-62856A, sc-62856B and sc-62856C.

RESEARCH USE

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

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

APPLICATIONS

PREP-2 siRNA (h) is recommended for the inhibition of PREP-2 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 μ M in 66 μ 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

PREP-2 (56.1): sc-101857 is recommended as a control antibody for monitoring of PREP-2 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 κ BP-HRP: sc-516102 or m-IgG κ 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 κ BP-FITC: sc-516140 or m-IgG κ 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 PREP-2 gene expression knockdown using RT-PCR Primer: PREP-2 (h)-PR: sc-62856-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

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