SANTA CRUZ BIOTECHNOLOGY, INC.

brachyury siRNA (h): sc-29820

BACKGROUND

The T-box gene family consists of members that share a unique DNA binding domain. The best characterized T-box (Tbx) gene, brachyury or T, encodes a transcription factor that plays an important role in early vertebrate development. Tbx genes are a family of developmental regulators with more than 20 members recently identified among invertebrates and vertebrates. Mutations in Tbx genes have been found to cause several human diseases. The understanding of functional mechanisms of Tbx products has come mainly from the prototypical T/brachyury protein, which is a transcription activator. The T-domain is a highly conserved DNA-binding motif originally defined in brachyury and characteristic of the Tbx family of transcription factors. The murine brachyury (T) gene is required in posterior mesoderm formation and axial development. Mutant embryos lacking T gene function are deficient in notochord differentiation and posterior mesoderm formation, but develop anterior mesoderm.

REFERENCES

1. Kispert, A., et al. 1994. Immunohistochemical analysis of the brachyury protein in wild-type and mutant mouse embryos. Dev. Biol. 161: 179-193.
2. Conlon, F.L., et al. 1995. Effects of the TWis mutation on notochord formation and mesodermal patterning. Mech. Dev. 49: 201-209.
3. Agulnik, S.I., et al. 1997. Three novel T-box genes in *Caenorhabditis elegans*. Genome 40: 458-464.
4. He, M.I., et al. 1999. Transcription repression by *Xenopus* ET and its human ortholog TBX3, a gene involved in ulnar-mammary syndrome. Proc. Natl. Acad. Sci. USA 96: 10212-10217.

CHROMOSOMAL LOCATION

Genetic locus: T (human) mapping to 6q27.

PRODUCT

brachyury 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 brachyury shRNA Plasmid (h): sc-29820-SH and brachyury shRNA (h) Lentiviral Particles: sc-29820-V as alternate gene silencing products.

For independent verification of brachyury (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-29820A, sc-29820B and sc-29820C.

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

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

brachyury (D-10): sc-166962 is recommended as a control antibody for monitoring of brachyury 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 brachyury gene expression knockdown using RT-PCR Primer: brachyury (h)-PR: sc-29820-PR (20 µl, 325 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Cerdan, C., et al. 2012. Activin A promotes hematopoietic fated mesoderm development through upregulation of brachyury in human embryonic stem cells. Stem Cells Dev. 21: 2866-2877.
2. Sun, S., et al. 2014. The T-box transcription factor brachyury promotes renal interstitial fibrosis by repressing E-cadherin expression. Cell Commun. Signal. 12: 76.
3. Shao, C., et al. 2015. The potential role of brachyury in inducing epithelial-to-mesenchymal transition (EMT) and HIF-1α expression in breast cancer cells. Biochem. Biophys. Res. Commun. 467: 1083-1089.
4. Li, K., et al. 2016. Brachyury promotes tamoxifen resistance in breast cancer by targeting SIRT1. Biomed. Pharmacother. 84: 28-33.

RESEARCH USE

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