SANTA CRUZ BIOTECHNOLOGY, INC.

OTX3 siRNA (h): sc-61271

BACKGROUND

Transcription factors, OTX1 and OTX2, are two murine homologs of the *Drosophila* orthodenticle (OTD), show a limited amino acid sequence divergence. OTX1 and OTX2 play an important role during early and later events required for proper brain development in that they are involved in the processes of induction, specification and regionalization of the brain. OTX1 is involved in corticogenesis, sensory organ development and pituitary functions, while OTX2 is necessary earlier in development, for the correct anterior neural plate specification and organization of the primitive streak. OTX2 is also required in the early specification of the neuroectoderm, which is destined to become the fore-midbrain, and both OTX1 and OTX2 cooperate in patterning the developing brain through a dosage-dependent mechanism. A related family member OTX3 is expressed in developing neural tissues and is required for postnatal survival, growth and brain development. OTX3 acts as a repressor of OTX2-mediated transactivation by forming a heterodimer with OTX2 on the TAATCC consensus motif.

REFERENCES

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2. Acampora, D., et al. 1999. Differential transcriptional control as the major molecular event in generating Otx1$^{-/-}$ and Otx2$^{-/-}$ divergent phenotypes. Development 126: 1417-1426.
3. Acampora, D., et al. 1999. Otx genes in corticogenesis and brain development. Cereb. Cortex 9: 533-542.
4. Acampora, D., et al. 1999. The TINS Lecture. Understanding the roles of Otx1 and Otx2 in the control of brain morphogenesis. Trends Neurosci. 22: 116-122.
5. Acampora, D., et al. 1999. Otx genes and the genetic control of brain morphogenesis. Mol. Cell. Neurosci. 13: 1-8.
6. Zhang, Y., et al. 2002. Identification, tissue expression, and a novel member of the Otx family. J. Biol. Chem. 277: 28065-28069.
7. Ohtoshi, A. and Behringer, R.R. 2004. Neonatal lethality, dwarfism, and abnormal brain development in Dmbx1 mutant mice. Mol. Cell. Biol. 24: 7548-7558.

CHROMOSOMAL LOCATION

Genetic locus: DMBX1 (human) mapping to 1p33.

PRODUCT

OTX3 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 OTX3 shRNA Plasmid (h): sc-61271-SH and OTX3 shRNA (h) Lentiviral Particles: sc-61271-V as alternate gene silencing products.

For independent verification of OTX3 (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-61271A, sc-61271B and sc-61271C.

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

OTX3 siRNA (h) is recommended for the inhibition of OTX3 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

OTX3 (E-9): sc-515294 is recommended as a control antibody for monitoring of OTX3 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 OTX3 gene expression knockdown using RT-PCR Primer: OTX3 (h)-PR: sc-61271-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.