

IFT172 siRNA (m): sc-146173

BACKGROUND

Intraflagellar transport is mediated by a variety of intraflagellar transport proteins (IFTs) that work in tandem to mediate ciliary and flagellar process assembly. Endogenous IFT proteins are most highly expressed within the inner segment, around the basal body, and within the outer segment. Additionally, IFT proteins are localized in discrete particles along the entire length of the axoneme. IFT proteins are divided into 2 subcomplexes, A and B, which contain at least 6 or 11 subunits, respectively. IFT-A proteins are associated with retrograde transport, whereas IFT-B proteins are thought to be involved in structure because, in their absence, cilia and flagella may be truncated, or completely absent. IFT172 (intraflagellar transport 172), also known as SLB, wim or osm-1, is a 1,749 amino acid protein that belongs to the IFT172 family and localizes to the cilium. Containing 14 TPR repeats and 9 WD repeats, IFT172 is required for the maintenance and formation of cilia. IFT172 plays an indirect role in Shh signaling, with cilia being required for all activity of the hedgehog pathway.

REFERENCES

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- Deane, J.A., et al. 2001. Localization of intraflagellar transport protein IFT52 identifies basal body transitional fibers as the docking site for IFT particles. *Curr. Biol.* 11: 1586-1590.
- Baker, S.A., et al. 2003. IFT20 links kinesin II with a mammalian intraflagellar transport complex that is conserved in motile flagella and sensory cilia. *J. Biol. Chem.* 278: 34211-34218.
- Huangfu, D., et al. 2003. Hedgehog signalling in the mouse requires intraflagellar transport proteins. *Nature* 426: 83-87.
- Tsujikawa, M. and Malicki, J. 2004. Intraflagellar transport genes are essential for differentiation and survival of vertebrate sensory neurons. *Neuron* 42: 703-716.
- Liu, A., et al. 2005. Mouse intraflagellar transport proteins regulate both the activator and repressor functions of Gli transcription factors. *Development* 132: 3103-3111.

CHROMOSOMAL LOCATION

Genetic locus: Ift172 (mouse) mapping to 5 B1.

PRODUCT

IFT172 siRNA (m) 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 IFT172 shRNA Plasmid (m): sc-146173-SH and IFT172 shRNA (m) Lentiviral Particles: sc-146173-V as alternate gene silencing products.

For independent verification of IFT172 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-146173A, sc-146173B and sc-146173C.

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

IFT172 siRNA (m) is recommended for the inhibition of IFT172 expression in mouse 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

IFT172 (A-11): sc-398393 is recommended as a control antibody for monitoring of IFT172 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 IFT172 gene expression knockdown using RT-PCR Primer: IFT172 (m)-PR: sc-146173-PR (20 μ 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 for detailed protocols and support products.