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Autotaxin siRNA (h): sc-44906

BACKGROUND

Autotaxin (ATX), also designated ectonucleotide pyrophosphatase/phosphodiesterase 2 (E-NPP 2), is a membrane-bound glycoprotein that cleaves diester bonds for a broad range of substrates. Originally isolated from the human melanoma cell line (A2058), Autotaxin is predominantly expressed in brain, placenta, ovary and small intestine. Autotaxin has significant homology to the cell membrane differentiation antigen PC-1, and is a stimulator of tumor cell motility. It also functions as a catalyst by hydrolytically removing 5'-nucleotides from the 3'-hydroxy termini of 3'-hydroxy-terminated oligonucleotides.

REFERENCES

1. Murata, J., et al. 1994. cDNA cloning of the human tumor motility-stimulating protein, Autotaxin, reveals a homology with phosphodiesterases. *J. Biol. Chem.* 269: 30479-30484.
2. Kawagoe, H., et al. 1995. Molecular cloning and chromosomal assignment of the human brain-type phosphodiesterase I/nucleotide pyrophosphatase gene (PDNP2). *Genomics* 30: 380-384.
3. Lee, H.Y., et al. 1996. Cloning, chromosomal localization, and tissue expression of Autotaxin from human teratocarcinoma cells. *Biochem. Biophys. Res. Commun.* 218: 714-719.
4. Mazereeuw-Hautier, J., et al. 2005. Production of lysophosphatidic acid in blister fluid: involvement of a lysophospholipase D activity. *J. Invest. Dermatol.* 125: 421-427.
5. Baumforth, K.R., et al. 2005. Induction of Autotaxin by the Epstein-Barr virus promotes the growth and survival of Hodgkin lymphoma cells. *Blood* 106: 2138-2146.

CHROMOSOMAL LOCATION

Genetic locus: ENPP2 (human) mapping to 8q24.12.

PRODUCT

Autotaxin 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 Autotaxin shRNA Plasmid (h): sc-44906-SH and Autotaxin shRNA (h) Lentiviral Particles: sc-44906-V as alternate gene silencing products.

For independent verification of Autotaxin (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-44906A, sc-44906B and sc-44906C.

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

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

Autotaxin (E-12): sc-374222 is recommended as a control antibody for monitoring of Autotaxin 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 Autotaxin gene expression knockdown using RT-PCR Primer: Autotaxin (h)-PR: sc-44906-PR (20 μ l, 597 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Iyer, P., et al. 2012. Autotaxin-lysophosphatidic acid axis is a novel molecular target for lowering intraocular pressure. *PLoS ONE* 7: e42627.
2. Ho, L.T.Y., et al. 2019. Role of the Autotaxin-lysophosphatidic acid axis in glaucoma, aqueous humor drainage and fibrogenic activity. *Biochim. Biophys. Acta Mol. Basis Dis.* 21: 165560.

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.