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TRAP siRNA (h): sc-44164

BACKGROUND

Tartrate-resistant acid phosphatase (TRAP, ACP5) is an iron containing glycoprotein that catalyzes the conversion of orthophosphoric monoester to alcohol and orthophosphate. TRAP is the most basic of the acid phosphatases and is the only form not inhibited by L⁺-tartrate. TRAP is a relatively minor lysosomal enzyme which may be activated in certain pathologies such as Hodgkin's disease and B- and T-cell leukemias. Receptor activator of NFκB ligand (RANKL) plays an essential role in osteoclast differentiation and activation by increasing the expression of protease osteoclast markers such as TRAP. TRAP has collagenolytic activity and plays a major role in ligament degradation.

REFERENCES

1. Fleckenstein, E., et al. 1996. Cloning and characterization of the human tartrate-resistant acid phosphatase (TRAP) gene. *Leukemia* 10: 637-643.
2. Fleckenstein, E.C., et al. 2000. The human tartrate-resistant acid phosphatase (TRAP): involvement of the hemin responsive elements (HRE) in transcriptional regulation. *Leuk. Lymphoma* 36: 603-612.
3. Capeller, B., et al. 2003. Evaluation of tartrate-resistant acid phosphatase (TRAP) 5b as serum marker of bone metastases in human breast cancer. *Anticancer Res.* 23: 1011-1015.
4. Wittrant Y., et al. 2003. Regulation of osteoclast protease expression by RANKL. *Biochem. Biophys. Res. Commun.* 310: 774-778.
5. Dwyer, K.W., et al. 2004. Blockade of the sympathetic nervous system degrades ligament in a rat MCL model. *J. Appl. Physiol.* 96: 711-718.
6. Nakano, Y., et al. 2004. Eccentric localization of osteocytes expressing enzymatic activities, protein, and mRNA signals for type 5 tartrate-resistant acid phosphatase (TRAP). *J. Histochem. Cytochem.* 52: 1475-1482.

CHROMOSOMAL LOCATION

Genetic locus: ACP5 (human) mapping to 19p13.2.

PRODUCT

TRAP 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 TRAP shRNA Plasmid (h): sc-44164-SH and TRAP shRNA (h) Lentiviral Particles: sc-44164-V as alternate gene silencing products.

For independent verification of TRAP (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-44164A, sc-44164B and sc-44164C.

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.

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

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

TRAP (D-3): sc-376875 is recommended as a control antibody for monitoring of TRAP 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 TRAP gene expression knockdown using RT-PCR Primer: TRAP (h)-PR: sc-44164-PR (20 μl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.