SANTA CRUZ BIOTECHNOLOGY, INC.

Phemx siRNA (h): sc-62798

BACKGROUND

Phemx (pan-hematopoietic expression protein), also known as PHMX, TSPAN32 (tetraspanin-32) or TSSC6 (tumor-suppressing subtransferable candidate 6), is a member of the tetraspanin (TM4SF) family of proteins that may be involved in transmembrane signal transduction, regulation of cell proliferation, differentiation and motility. Phemx is a multi-pass membrane protein containing intracellular N- and C-terminal domains, four transmembrane domains and two extracellular loops. It is ubiquitously expressed from early embryogenesis through adulthood. Phemx exhibits predominant expression in hematopoietic tissues suggesting a role in hematopoietic-cell function. In association with the Integrin αIIb/Integrin β3 complex, Phemx functions to stabilize arterial thrombi in platelets and regulate "outside-in" signaling. This interaction may be important in the process of wound healing. The gene encoding Phemx is located in an important tumor-suppressor gene region that has been associated with Beckwith-Wiedemann syndrome as well as a variety of cancers.

REFERENCES

1. Lee, M.P., et al. 1999. Two novel genes in the center of the 11p15 imprinted domain escape genomic imprinting. Hum. Mol. Genet. 8: 683-690.
2. Nicholson, R.H., et al. 2000. Phemx, a novel mouse gene expressed in hematopoietic cells maps to the imprinted cluster on distal chromosome 7. Genomics 68: 13-21.
3. Paulsen, M., et al. 2000. Sequence conservation and variability of imprinting in the Beckwith-Wiedemann syndrome gene cluster in human and mouse. Hum. Mol. Genet. 9: 1829-1841.
4. Harada, Y., et al. 2001. A hematopoietic-specific transmembrane protein, Art-1, is possibly regulated by AML1. Biochem. Biophys. Res. Commun. 284: 714-722.
5. Robb, L., et al. 2001. Molecular characterisation of mouse and human TSSC6: evidence that TSSC6 is a genuine member of the tetraspanin superfamily and is expressed specifically in haematopoietic organs. Biochim. Biophys. Acta 1522: 31-41.
6. Tarrant, J.M., et al. 2002. The absence of Tssc6, a member of the tetraspanin superfamily, does not affect lymphoid development but enhances *in vitro* T-cell proliferative responses. Mol. Cell. Biol. 22: 5006-5018.

CHROMOSOMAL LOCATION

Genetic locus: TSPAN32 (human) mapping to 11p15.5.

PRODUCT

Phemx 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 Phemx shRNA Plasmid (h): sc-62798-SH and Phemx shRNA (h) Lentiviral Particles: sc-62798-V as alternate gene silencing products.

For independent verification of Phemx (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-62798A, sc-62798B and sc-62798C.

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

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

Phemx (FF-37): sc-81997 is recommended as a control antibody for monitoring of Phemx 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 Phemx gene expression knockdown using RT-PCR Primer: Phemx (h)-PR: sc-62798-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.