

Pilt siRNA (m): sc-152265

BACKGROUND

Pilt (protein incorporated later into tight junctions), also known as TJAP1 (tight junction associated protein 1 (peripheral)) or TJP4 (tight junction protein 4 (peripheral)), is a 557 amino acid phosphoprotein that contains an N-terminal coiled-coil domain and a central proline-rich region. Encoded by a gene that maps to human chromosome 6p21.1, Pilt shares 74% amino acid identity with its mouse homolog and exists as four alternatively spliced isoforms. Ubiquitously expressed, Pilt localizes to Golgi apparatus and may function in vesicle trafficking. Pilt interacts with SAP 97 and also binds PSD-93. Although Pilt is recruited to tight junctions (TJ) during late stages of TJ complex maturation, it is excluded from apical junctions and desmosomes. Pilt may function as a heart-rhythm-determinant (HRD) gene that plays a role in sex-dependent organizational principles of the heart-rhythm transcriptome.

REFERENCES

1. Kawabe, H., Nakanishi, H., Asada, M., Fukuhara, A., Morimoto, K., Takeuchi, M. and Takai, Y. 2001. Pilt, a novel peripheral membrane protein at tight junctions in epithelial cells. *J. Biol. Chem.* 276: 48350-48355.
2. Yuen, T., Ruf, F., Chu, T. and Sealfon, S.C. 2009. Microtranscriptome regulation by gonadotropin-releasing hormone. *Mol. Cell. Endocrinol.* 302: 12-17.
3. Si, H., Banga, R.S., Kapitsinou, P., Ramaiah, M., Lawrence, J., Kambhampati, G., Gruenwald, A., Bottinger, E., Glicklich, D., Tellis, V., Greenstein, S., Thomas, D.B., Pullman, J., Fazzari, M. and Susztak, K. 2009. Human and murine kidneys show gender- and species-specific gene expression differences in response to injury. *PLoS ONE* 4: e4802.
4. Iacobas, D.A., Iacobas, S., Thomas, N. and Spray, D.C. 2010. Sex-dependent gene regulatory networks of the heart rhythm. *Funct. Integr. Genomics* 10: 73-86.
5. Wang, X.R., Zhang, X.M., Zhen, J., Zhang, P.X., Xu, G. and Jiang, H. 2010. MicroRNA expression in the embryonic mouse inner ear. *Neuroreport* 21: 611-617.
6. Knowlton, M.L., Selfors, L.M., Wrobel, C.N., Gu, T.L., Ballif, B.A., Gygi, S.P., Polakiewicz, R. and Brugge, J.S. 2010. Profiling Y561-dependent and -independent substrates of CSF-1R in epithelial cells. *PLoS ONE* 5: e13587.

CHROMOSOMAL LOCATION

Genetic locus: Tjp1 (mouse) mapping to 17 C.

PRODUCT

Pilt siRNA (m) is a pool of 2 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 Pilt shRNA Plasmid (m): sc-152265-SH and Pilt shRNA (m) Lentiviral Particles: sc-152265-V as alternate gene silencing products.

For independent verification of Pilt (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-152265A and sc-152265B.

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

Pilt siRNA (m) is recommended for the inhibition of Pilt 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Pilt gene expression knockdown using RT-PCR Primer: Pilt (m)-PR: sc-152265-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.