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# ZO-1 siRNA (h2): sc-44288

## BACKGROUND

Tight junctions are complexes of proteins that create intercellular boundaries between the plasma membrane domains of epithelial and endothelial cells. Many of the tight junction-associated proteins are members of the membrane-associated guanylate kinase (MAGUK) family and include occludin, ZO-1, ZO-2 and ZO-3. These proteins are thought to have both structural and signaling roles, and are characteristically defined by three protein-protein interaction modules: the PDZ domain, the SH3 domain and the guanylate kinase (GuK) domain. ZO-1 forms complexes with either ZO-2 or ZO-3. In addition, these proteins can also associate with claudin, Occludin and F-Actin, at tight junction stands, where they provide a linkage between the Actin cytoskeleton and the tight junction. ZO-1 expression is significantly reduced in many breast cancer lines. ZO-2 and ZO-3 are ubiquitously expressed within epithelial tight junctions and, unlike ZO-1, which is also expressed at cell junctions of cardiac myocytes, ZO-2 is not expressed in nonepithelial tissue.

## REFERENCES

1. Furuse, M., et al. 1994. Direct association of Occludin with ZO-1 and its possible involvement in the localization of Occludin at tight junctions. *J. Cell Biol.* 127: 1617-1626.
2. Anderson, J.M. 1996. Cell signalling: MAGUK magic. *Curr. Biol.* 6: 382-384.
3. Hoover, K.B., et al. 1998. Loss of the tight junction MAGUK ZO-1 in breast cancer: relationship to glandular differentiation and loss of heterozygosity. *Am. J. Pathol.* 153: 1767-1773.
4. Haskins, J., et al. 1998. ZO-3, a novel member of the MAGUK protein family found at the tight junction, interacts with ZO-1 and occludin. *J. Cell Biol.* 141: 199-208.
5. Itoh, M., et al. 1999. Direct binding of three tight junction-associated MAGUKs, ZO-1, ZO-2, and ZO-3, with the COOH termini of claudins. *J. Cell Biol.* 147: 1351-1363.

## CHROMOSOMAL LOCATION

Genetic locus: TJP1 (human) mapping to 15q13.1.

## PRODUCT

ZO-1 siRNA (h2) 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see ZO-1 shRNA Plasmid (h2): sc-44288-SH and ZO-1 shRNA (h2) Lentiviral Particles: sc-44288-V as alternate gene silencing products.

For independent verification of ZO-1 (h2) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-44288A, sc-44288B and sc-44288C.

## PROTOCOLS

See our web site at [www.scbt.com](http://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  $\mu$ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330  $\mu$ l of RNase-free water makes a 10  $\mu$ M solution in a 10  $\mu$ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

## APPLICATIONS

ZO-1 siRNA (h2) is recommended for the inhibition of ZO-1 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  $\mu$ M in 66  $\mu$ 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

ZO-1 (R40.76): sc-33725 is recommended as a control antibody for monitoring of ZO-1 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).

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor ZO-1 gene expression knockdown using RT-PCR Primer: ZO-1 (h2)-PR: sc-44288-PR (20  $\mu$ l, 540 bp). 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.