SANTA CRUZ BIOTECHNOLOGY, INC.

ZFYVE16 siRNA (h): sc-91838

BACKGROUND

Zinc-finger proteins contain DNA-binding domains and have a wide variety of functions, most of which encompass some form of transcriptional activation or repression. ZFYVE16 (zinc finger FYVE domain-containing protein 16), also known as endofin or endosome-associated FYVE domain protein, is a 1,539 amino acid protein containing one FYVE-type zinc finger domain. This domain, which has been shown to be necessary for the localization of ZFYVE16 to the early endosomes, mediates the association of ZFYVE16 with phosphatidylinositol 3-phosphate (PI3P). As a membrane-associated protein, ZFYVE16 also interacts with Smad4 and TOM1 to regulate membrane trafficking in the endosomal pathway. Three named isoforms of ZFYVE16 exist as a result of alternative splicing events.

REFERENCES

1. Hayes, S., et al. 2002. TGF β receptor internalization into EEA1-enriched early endosomes: role in signaling to Smad2. J. Cell Biol. 158: 1239-1249.
2. Seet, L.F., et al. 2004. Endofin recruits TOM1 to endosomes. J. Biol. Chem. 279: 4670-4679.
3. Runyan, C.E., et al. 2005. The role of internalization in transforming growth factor β1-induced Smad2 association with Smad anchor for receptor activation (SARA) and Smad2-dependent signaling in human mesangial cells. J. Biol. Chem. 280: 8300-8308.
4. Seet, L.F., et al. 2005. Endofin recruits clathrin to early endosomes via TOM1. J. Cell Sci. 118: 575-587.
5. Chen, Y.G., et al. 2007. Endofin, a FYVE domain protein, interacts with Smad4 and facilitates transforming growth factor-β signaling. J. Biol. Chem. 282: 9688-9695.
6. Shi, W., et al. 2007. Endofin acts as a Smad anchor for receptor activation in BMP signaling. J. Cell Sci. 120: 1216-1224.
7. Chen, Y., et al. 2007. Phosphoproteomics identified Endofin, DCBLD2, and KIAA0582 as novel tyrosine phosphorylation targets of EGF signaling and Iressa in human cancer cells. Proteomics 7: 2384-2397.

CHROMOSOMAL LOCATION

Genetic locus: ZFYVE16 (human) mapping to 5q14.1.

PRODUCT

ZFYVE16 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 ZFYVE16 shRNA Plasmid (h): sc-91838-SH and ZFYVE16 shRNA (h) Lentiviral Particles: sc-91838-V as alternate gene silencing products.

For independent verification of ZFYVE16 (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-91838A, sc-91838B and sc-91838C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

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

ZFYVE16 siRNA (h) is recommended for the inhibition of ZFYVE16 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

ZFYVE16 (1H6): sc-135608 is recommended as a control antibody for monitoring of ZFYVE16 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 ZFYVE16 gene expression knockdown using RT-PCR Primer: ZFYVE16 (h)-PR: sc-91838-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.