SANTA CRUZ BIOTECHNOLOGY, INC.

PI 3-kinase p100 siRNA (m): sc-62803

BACKGROUND

Phosphatidylinositol 3-kinases (PI3Ks) phosphorylate the 3' OH position of the inositol ring of inositol lipids. PI 3-kinase p100 (phosphoinositide-3-kinase p100 subunit), also known as hVps34 or PIK3C3 (phosphoinositide-3-kinase, class III), is a member of the PI 3/PI 4-kinase family. It is ubiquitously expressed with predominant expression in skeletal muscle and is believed to participate in endosome to lysosome transport, multivesicular body formation, autophagy and retrograde endosome to Golgi transport. PI 3-kinase p100 is the catalytic subunit of class III PI3Ks and forms a heterodimer with p150, a regulatory subunit of class 3 PI3Ks. PI 3-kinase p100 exclusively phosphorylates phosphatidylinositol to produce PtdIns3P. Unlike class I PI3Ks, whose activities are enhanced in the presence of magnesium, PI 3-kinase p100 activity is enhanced by manganese. Its activity can also be regulated by nutrients, suggesting an important role of PI-3 kinase p100 in the regulation of mTOR protein synthesis and autophagy.

REFERENCES

1. Volinia, S., et al. 1995. A human phosphatidylinositol 3-kinase complex related to the yeast Vps34p-Vps15p protein sorting system. EMBO J. 14: 3339-3348.
2. Stopkova, P., et al. 2004. Identification of PIK3C3 promoter variant associated with bipolar disorder and schizophrenia. Biol. Psychiatry 55: 981-988.
3. Furuya, N., et al. 2005. The evolutionarily conserved domain of Beclin 1 is required for Vps34 binding, autophagy and tumor suppressor function. Autophagy 1: 46-52.
4. Hall, B.S., et al. 2006. TbVps34, the trypanosome orthologue of Vps34, is required for Golgi complex segregation. J. Biol. Chem. 281: 27600-27612.
5. Chu, C.T., et al. 2007. Beclin 1-independent pathway of damage-induced mitophagy and autophagic stress: implications for neurodegeneration and cell death. Autophagy 3: 663-666.

CHROMOSOMAL LOCATION

Genetic locus: Pik3c3 (mouse) mapping to 18 B1.

PRODUCT

PI 3-kinase p100 siRNA (m) 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 PI 3-kinase p100 shRNA Plasmid (m): sc-62803-SH and PI 3-kinase p100 shRNA (m) Lentiviral Particles: sc-62803-V as alternate gene silencing products.

For independent verification of PI 3-kinase p100 (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-62803A, sc-62803B and sc-62803C.

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

PI 3-kinase p100 siRNA (m) is recommended for the inhibition of PI 3-kinase p100 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 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

PI 3-kinase p100 (F-11): sc-365404 is recommended as a control antibody for monitoring of PI 3-kinase p100 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 PI 3-kinase p100 gene expression knockdown using RT-PCR Primer: PI 3-kinase p100 (m)-PR: sc-62803-PR (20 µl, 562 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.