SANTA CRUZ BIOTECHNOLOGY, INC.

ILKAP siRNA (m): sc-146223

BACKGROUND

ILKAP (integrin-linked kinase-associated serine/threonine phosphatase 2C), also known as PP2Cδ, is a 392 amino acid cytoplasmic protein phosphatase that selectively interacts with integrin linked kinase (ILK) to regulate growth factor signaling and cell adhesion. While widely expressed, ILKAP is found at highest levels in striated muscle with lower levels found in smooth muscle. ILKAP belongs to the PP2C family and contains one PP2C-like domain. ILKAP has been suggested to inhibit oncogenic transformation and the ILK-GSK3β signaling axis, and can bind two magnesium or manganese ions per subunit as cofactors. The gene encoding ILKAP maps to human chromosome 2, which consists of 237 million bases encoding over 1,400 genes and making up approximately 8% of the human genome.

REFERENCES

1. Tong, Y., et al. 1998. Cloning and characterization of a novel mammalian PP2C isozyme. J. Biol. Chem. 273: 35282-35290.
2. Leung-Hagesteijn, C., et al. 2001. Modulation of integrin signal transduction by ILKAP, a protein phosphatase 2C associating with the integrin-linked kinase, ILK1. EMBO J. 20: 2160-2170.
3. Kumar, A.S., et al. 2004. ILKAP regulates ILK signaling and inhibits anchorage-independent growth. Oncogene 23: 3454-3461.
4. Tamura, S., et al. 2006. PP2C family members play key roles in regulation of cell survival and apoptosis. Cancer Sci. 97: 563-567.
5. Lammers, T. and Lavi, S. 2007. Role of type 2C protein phosphatases in growth regulation and in cellular stress signaling. Crit. Rev. Biochem. Mol. Biol. 42: 437-461.
6. Nakrieko, K.A., et al. 2008. Modulation of integrin-linked kinase nucleocytoplasmic shuttling by ILKAP and CRM1. Cell Cycle 7: 2157-2166.
7. Pridgeon, J.W., et al. 2009. Proteomic analysis reveals Hrs ubiquitin-interacting motif-mediated ubiquitin signaling in multiple cellular processes. FEBS J. 276: 118-131.

CHROMOSOMAL LOCATION

Genetic locus: Ilkap (mouse) mapping to 1 D.

PRODUCT

ILKAP 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 ILKAP shRNA Plasmid (m): sc-146223-SH and ILKAP shRNA (m) Lentiviral Particles: sc-146223-V as alternate gene silencing products.

For independent verification of ILKAP (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-146223A, sc-146223B and sc-146223C.

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

ILKAP siRNA (m) is recommended for the inhibition of ILKAP 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

ILKAP (41): sc-136341 is recommended as a control antibody for monitoring of ILKAP 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 ILKAP gene expression knockdown using RT-PCR Primer: ILKAP (m)-PR: sc-146223-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.