SANTA CRUZ BIOTECHNOLOGY, INC.

Ska2 siRNA (h): sc-94176

BACKGROUND

Ska2 (spindle and kinetochore associated complex subunit 2), also known as FAM33A, is a 121 amino acid component of the Ska1 complex, a microtubule-binding subcomplex of the outer kinetochore that is critical for proper chromosome segregation. The Ska1 complex is a component of the kinetochore-microtubule interface and directly associates with microtubules as oligomeric assemblies. Localized to the outer kinetochore and spindle microtubules during cell proliferation, Ska2 is essential for spindle checkpoint silencing and exit from mitosis. Downregulation of Ska2 leads to delayed recruitment of MAD2, a component of the mitotic spindle checkpoint, to several kinetochores resulting in occasional loss of individual chromosomes from the metaphase plate. Ska2 is encoded by a gene located on human chromosome 17, which comprises over 2.5% of the human genome and encodes over 1,200 genes.

REFERENCES

1. Hanisch, A., et al. 2006. Timely anaphase onset requires a novel spindle and kinetochore complex comprising Ska1 and Ska2. EMBO J. 25: 5504-5515.
2. Wang, C. and St Leger, R.J. 2007. The MAD1 adhesin of *Metarhizium anisopliae* links adhesion with blastospore production and virulence to insects, and the MAD2 adhesin enables attachment to plants. Eukaryotic Cell 6: 808-816.
3. Rice, L., et al. 2008. Identification and functional analysis of SKA2 interaction with the glucocorticoid receptor. J. Endocrinol. 198: 499-509.
4. Daub, H., et al. 2008. Kinase-selective enrichment enables quantitative phosphoproteomics of the kinome across the cell cycle. Mol. Cell 31: 438-448.
5. Daum, J.R., et al. 2009. Ska3 is required for spindle checkpoint silencing and the maintenance of chromosome cohesion in mitosis. Curr. Biol. 19: 1467-1472.
6. Welburn, J.P., et al. 2009. The human kinetochore Ska1 complex facilitates microtubule depolymerization-coupled motility. Dev. Cell 16: 374-385.
7. Guimaraes, G.J. and Deluca, J.G. 2009. Connecting with Ska, a key complex at the kinetochore-microtubule interface. EMBO J. 28: 1375-1377.

CHROMOSOMAL LOCATION

Genetic locus: SKA2 (human) mapping to 17q22.

PRODUCT

Ska2 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 Ska2 shRNA Plasmid (h): sc-94176-SH and Ska2 shRNA (h) Lentiviral Particles: sc-94176-V as alternate gene silencing products.

For independent verification of Ska2 (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-94176A, sc-94176B and sc-94176C.

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

Ska2 siRNA (h) is recommended for the inhibition of Ska2 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

Ska2 (H-2): sc-514495 is recommended as a control antibody for monitoring of Ska2 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 Ska2 gene expression knockdown using RT-PCR Primer: Ska2 (h)-PR: sc-94176-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.

PROTOCOLS

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