SANTA CRUZ BIOTECHNOLOGY, INC.

Spot 14 siRNA (h): sc-63058

BACKGROUND

Spot 14, also known as S14 or THRSP (thyroid hormone responsive protein), is a small acidic protein localizing to the nucleus and can be found in tissues that synthesize triglycerides, such as liver, mammary glands and adipose tissues. Spot 14 is implicated in growth and differentiation, possibly functioning as a transcription regulator for genes encoding proteins that participate in lipogenesis. A variety of lipogenic stimuli can activate the expression of Spot 14, including thyroid hormone, dietary carbohydrate, Insulin and glucose. Its expression can be downregulated by catecholamine and glucagon. In addition, Spot 14 expression is known to oscillate with the circadian clock. Knockdown of Spot 14 leads to impaired lipid synthesis and apoptosis. In most breast cancers, Spot 14 is overexpressed and is believed to augment cell growth and survival.

REFERENCES

1. Grillasca, J.P., et al. 1997. Cloning and initial characterization of human and mouse Spot 14 genes. FEBS Lett. 401: 38-42.
2. Cunningham, B.A., et al. 1998. "Spot 14" protein: a metabolic integrator in normal and neoplastic cells. Thyroid 8: 815-825.
3. Moncur, J.T., et al. 1998. The "Spot 14" gene resides on the telomeric end of the 11q13 amplicon and is expressed in lipogenic breast cancers: implications for control of tumor metabolism. Proc. Natl. Acad. Sci. USA 95: 6989-6994.
4. Compe, E., et al. 2001. Spot 14 protein interacts and cooperates with chicken ovalbumin upstream promoter-transcription factor 1 in the transcription of the L-type pyruvate kinase gene through a specificity protein 1 (Sp1) binding site. Biochem. J. 358: 175-183.
5. Sanchez-Rodriguez, J., et al. 2005. The Spot 14 protein inhibits growth and induces differentiation and cell death of human MCF7 breast cancer cells. Biochem. J. 390: 57-65.
6. Kinlaw, W.B., et al. 2006. Spot 14: A marker of aggressive breast cancer and a potential therapeutic target. Endocrinology 147: 4048-4055.
7. Wells, W.A., et al. 2006. Expression of "Spot 14" (THRSP) predicts disease free survival in invasive breast cancer: immunohistochemical analysis of a new molecular marker. Breast Cancer Res. Treat. 98: 231-240.

CHROMOSOMAL LOCATION

Genetic locus: THRSP (human) mapping to 11q14.1.

PRODUCT

Spot 14 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 Spot 14 shRNA Plasmid (h): sc-63058-SH and Spot 14 shRNA (h) Lentiviral Particles: sc-63058-V as alternate gene silencing products.

For independent verification of Spot 14 (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-63058A, sc-63058B and sc-63058C.

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

Spot 14 siRNA (h) is recommended for the inhibition of Spot 14 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

Spot 14 (F-7): sc-137178 is recommended as a control antibody for monitoring of Spot 14 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 Spot 14 gene expression knockdown using RT-PCR Primer: Spot 14 (h)-PR: sc-63058-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.