SANTA CRUZ BIOTECHNOLOGY, INC.

FBL10 siRNA (h): sc-75005

BACKGROUND

FBXL10 (F-box and leucine-rich repeat protein 10), also known as FBL10, CXXC2 (CXXC-type zinc finger protein 2), PCCX2, KDM2B, JEMMA (jumonji domain, EMSY-interactor, methyltransferase motif protein) or JHDM1B (jumonji C domain-containing histone demethylase 1B), is a nuclear protein that contains one F-box domain, a CXXC-type zinc finger, one JMJC domain, four leucine-rich repeats and one PHD-type zinc finger. FBXL10 belongs to the Fbls class of F-box proteins that contain leucine-rich repeats in addition to their F-box motif. F-box proteins are critical components of the SCF (Skp1-CUL-1-F-box protein) type E3 ubiquitin ligase complex and are involved in substrate recognition and recruitment for ubiquitination. In addition to its role in substrate recognition as a component of the E3 complex, FBXL10 functions as a dimethylation-specific demethylase, binding iron as a cofactor and demethylating lysine-36 of Histone H3. This suggests that FBXL10 plays a central role in the histone code.

REFERENCES

1. Winston, J.T., et al. 1999. A family of mammalian F-box proteins. Curr. Biol. 9: 1180-1182.
2. Fujino, T., et al. 2000. PCCX1, a novel DNA-binding protein with PHD finger and CXXC domain, is regulated by proteolysis. Biochem. Biophys. Res. Commun. 271: 305-310.
3. Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 609078. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/
4. Jin, J., et al. 2004. Systematic analysis and nomenclature of mammalian F-box proteins. Genes Dev. 18: 2573-2580.
5. Suzuki, T., et al. 2006. Tumor suppressor gene identification using retroviral insertional mutagenesis in Blm-deficient mice. EMBO J. 25: 3422-3431.
6. Tsukada, Y., et al. 2006. Histone demethylation by a family of JmjC domain-containing proteins. Nature 439: 811-816.
7. Frescas, D., et al. 2007. JHDM1B/FBXL10 is a nucleolar protein that represses transcription of ribosomal RNA genes. Nature 450: 309-313.
8. Koyama-Nasu, R., et al. 2007. The F-box protein Fbl10 is a novel transcriptional repressor of c-Jun. Nat. Cell Biol. 9: 1074-1080.

CHROMOSOMAL LOCATION

Genetic locus: KDM2B (human) mapping to 12q24.31.

PRODUCT

FBL10 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 FBL10 shRNA Plasmid (h): sc-75005-SH and FBL10 shRNA (h) Lentiviral Particles: sc-75005-V as alternate gene silencing products.

For independent verification of FBL10 (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-75005A, sc-75005B and sc-75005C.

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

FBL10 siRNA (h) is recommended for the inhibition of FBL10 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

FBL10 (5G1): sc-293279 is recommended as a control antibody for monitoring of FBL10 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 FBL10 gene expression knockdown using RT-PCR Primer: FBL10 (h)-PR: sc-75005-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.