SANTA CRUZ BIOTECHNOLOGY, INC.

FBXO3 siRNA (h): sc-96506

BACKGROUND

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. They are members of a larger family of proteins that are involved in the regulation of a wide variety of cellular processes (including the cell cycle, immune responses, signaling cascades and developmental events) through the targeting of proteins, such as cyclins, cyclin-dependent kinase inhibitors, IκB-α and β-catenin, for proteasomal degradation. FBXO3 (F-box protein 3), also known as FBA or FBX3, is a 471 amino acid member of the F-box protein family. Substrate-recognition component of the SCF (SKP1-CUL1-F-box protein)-type E3 ubiquitin ligase complex, FBXO3 contains an apaG domain and a F-box domain. Existing as two isoforms producted by alternative splicing events, FBXO3 interacts with Skp1 p19 and CUL-1.

REFERENCES

1. Bai, C., et al. 1996. SKP1 connects cell cycle regulators to the ubiquitin proteolysis machinery through a novel motif, the F-box. Cell 86: 263-274.
2. Cenciarelli, C., et al. 1999. Identification of a family of human F-box proteins. Curr. Biol. 9: 1177-1179.
3. Winston, J.T., et al. 1999. A family of mammalian F-box proteins. Curr. Biol. 9: 1180-1182.
4. Latres, E., et al. 1999. The human F box protein β-Trcp associates with the Cul1/Skp1 complex and regulates the stability of β-catenin. Oncogene 18: 849-854.
5. Masuda, K., et al. 2002. Molecular profile of synovial fibroblasts in rheumatoid arthritis depends on the stage of proliferation. Arthritis Res. 4: R8.
6. Ilyin, G.P., et al. 2002. A new subfamily of structurally related human F-box proteins. Gene 296: 11-20.
7. Yoshida, Y., et al. 2002. E3 ubiquitin ligase that recognizes sugar chains. Nature 418: 438-442.
8. Shima, Y., et al. 2008. PML activates transcription by protecting HIPK2 and p300 from SCFFbx3-mediated degradation. Mol. Cell. Biol. 28: 7126-7138.

CHROMOSOMAL LOCATION

Genetic locus: FBXO3 (human) mapping to 11p13.

PRODUCT

FBXO3 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 FBXO3 shRNA Plasmid (h): sc-96506-SH and FBXO3 shRNA (h) Lentiviral Particles: sc-96506-V as alternate gene silencing products.

For independent verification of FBXO3 (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-96506A, sc-96506B and sc-96506C.

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

FBXO3 siRNA (h) is recommended for the inhibition of FBXO3 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

FBXO3 (C-7): sc-514625 is recommended as a control antibody for monitoring of FBXO3 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker™ compatible goat anti-mouse IgG-HRP: sc-2031 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor FBXO3 gene expression knockdown using RT-PCR Primer: FBXO3 (h)-PR: sc-96506-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.