β -TrCP siRNA (m): sc-37179



The Power to Question

BACKGROUND

 $\beta\text{-TrCP}$ (beta-tranducin repeats containing protein), also designated E3RSIkB or FWD1, and HOS (homologous to Slimb) are F-box proteins that function as substrate recognition subunits of ubiquitin ligases. HOS and $\beta\text{-TrCP}$ differ in their amino terminal regions, but exhibit high homology within the F-box and WD40 repeat-containing regions. $\beta\text{-TrCP}$ mediates ubiquitin/ proteasome-dependent degradation of CD4 and ubiquitination of various proteins including IkB and $\beta\text{-catenin}$. HOS has also been shown to regulate the degradation of IkB and $\beta\text{-catenin}$ in a similar manner.

REFERENCES

- 1. Hatakeyama, S., et al. 1990. Ubiquitin-dependent degradation of $l\kappa B\alpha$ is mediated by a ubiquitin ligase Skp1/Cul 1/F-box protein FWD1. Proc. Natl. Acad. Sci. USA 96: 3859-3863.
- Margottin, F., et al. 1998. A novel human WD protein, h-β TrCp, that interacts with HIV-1 Vpu connects CD4 to the ER degradation pathway through an F-box motif. Mol. Cell 1: 565-574.
- 3. Yaron, A., et al. 1998. Identification of the receptor component of the $l\kappa B\alpha$ -ubiquitin ligase. Nature 396: 590-594.
- Zhou, P., et al. 1998. Ubiquitination and degradation of the substrate recognition subunits of SCF ubiquitin-protein ligases. Mol. Cell 2: 571-580.
- 5. Fuchs, S.Y., et al. 1999. HOS, a human homolog of Slimb, forms an SCF complex with Skp1 and Cullin1 and targets the phosphorylation-dependent degradation of lkB and β -catenin. Oncogene 18: 2039-2046.

CHROMOSOMAL LOCATION

Genetic locus: BTRC (human) mapping to 10q25; Btrc (mouse) mapping to 19 D1.

PRODUCT

 $\beta\text{-TrCP}$ siRNA (m) is a pool of 3 target-specific 20-25 nt siRNAs designed to knock down gene expression. Each vial contains 3 nmol of lyophilized siRNA, sufficient for a 10 μM solution once resuspended using protocol below. Suitable for 50-100 transfections.

For independent verification of β -TrCP (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3 nmol of lyophilized siRNA. These include: sc-37179A, sc-37179B and sc-37179C.

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 μ l of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNAse-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

 $\beta\text{-TrCP}$ siRNA (m) is recommended for the inhibition of $\beta\text{-TrCP}$ expression in mouse cells.

β-TrCP (H-300): sc-15354 is recommended as a control antibody for Western Blotting (starting dilution 1:100, dilution range 1:100-1:1,000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500) protein detection using the recommended secondary reagents listed below.

RECOMMENDED SECONDARY REAGENTS

To ensure optimal results, the following support (secondary) reagents are recommended: 1) Western Blotting: use goat anti-rabbit IgG-HRP: sc-2004 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible goat anti-rabbit IgG-HRP: sc-2030 (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) Immunofluo-rescence: use goat anti-rabbit IgG-FITC: sc-2012 (dilution range: 1:100-1:400) or goat anti-rabbit IgG-TR: sc-2780 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

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 10µM in 60 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. Semi-quantitative RT-PCR may be performed using RT-PCR Primer: β -TrCP (m)-PR: sc-37179-PR (20 µl, 431 bp). 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 or our catalog for detailed protocols and support products.

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