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SANTA CRUZ BIOTECHNOLOGY, INC.

CHIP siRNA (h): sc-43555

BACKGROUND

CHIP (carboxy terminus of HSP 70-interacting protein), also designated STIP1 homology and U-box containing protein 1, HSPABP2, NY-CO-7, SDCCAG7 and STUB1, is a cytoplasmic E3 ubiquitin ligase that influences protein ubiquitylation. CHIP interacts with Smad1/Smad4 and blocks BMP signaling through the ubiquitin-mediated degradation of Smad proteins. CHIP controls both association of HSP70/Hsp90 chaperones with ErbB2 and down-regulation of ErbB2 induced by inhibitors of Hsp90. A 1.3-kb transcript is most abundant in striated muscle (heart and skeletal muscle), with lower expression in pancreas and brain.

CHROMOSOMAL LOCATION

Genetic locus: STUB1 (human) mapping to 16p13.3.

PRODUCT

CHIP 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 CHIP shRNA Plasmid (h): sc-43555-SH and CHIP shRNA (h) Lentiviral Particles: sc-43555-V as alternate gene silencing products.

For independent verification of CHIP (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-43555A, sc-43555B and sc-43555C.

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

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

CHIP (G-2): sc-133066 is recommended as a control antibody for monitoring of CHIP 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 CHIP gene expression knockdown using RT-PCR Primer: CHIP (h)-PR: sc-43555-PR (20 µl, 416 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Jang, K.W., et al. 2011. The C-terminus of HSP70-interacting protein promotes Met receptor degradation. J. Thorac. Oncol. 6: 679-687.
2. Lim, J.H. and Woo, C.H. 2011. Laminar flow activation of ERK5 leads to cytoprotective effect via CHIP-mediated p53 ubiquitination in endothelial cells. Anat. Cell Biol. 44: 265-273.
3. Wang, J., et al. 2011. Gambogic acid-induced degradation of mutant p53 is mediated by proteasome and related to CHIP. J. Cell. Biochem. 112: 509-519.
4. Jang, K.W., et al. 2011. Ubiquitin ligase CHIP induces TRAF2 proteasomal degradation and NFκB inactivation to regulate breast cancer cell invasion. J. Cell. Biochem. 112: 3612-3620.
5. Blessing, N.A., et al. 2014. The E3 ligase CHIP mediates ubiquitination and degradation of mixed-lineage kinase 3. Mol. Cell. Biol. 34: 3132-3143.
6. Yong, H.J., et al. 2017. Von Hippel-Lindau regulates interleukin-32β stability in ovarian cancer cells. Oncotarget 8: 69833-69846.
7. Cho, S.H., et al. 2017. The antitumor effect of C-terminus of HSP70-interacting protein via degradation of c-Met in small cell lung cancer. Korean J. Thorac. Cardiovasc. Surg. 50: 153-162.

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.