SANTA CRUZ BIOTECHNOLOGY, INC.

RING1B siRNA (h): sc-62946

BACKGROUND

Polycomb group (PcG) proteins form multiprotein complexes that regulate expression patterns of developmental and cell proliferation genes. RING1B (E3 ubiquitin-protein ligase RING2), also known as RING2, RING finger protein BAP-1, DinG protein or HIP2-interacting protein 3, is a PcG protein involved in protein degradation and ubiquitination pathways. As an E3 ubiquitin-protein ligase that mediates monoubiquination of Histone H2A, RING1B is an essential component of the chromatin-associated class II PcG repressive complex 1 (PRC1/hPRC-H), a complex that represses transcription of many genes throughout development. RING1B contains one RING-type zinc finger domain that interacts with Huntington interacting protein 2 (HIP2) within the PRC1 complex during E3 ubiquitin ligase activity. Subcellularly located in the nucleus, RING1B may be involved in random and imprinted inactivation of the X chromosome in female mammals. Underexpression of RING1B in mice cause axial skeletal abnormalities and reduced expression of some HOX genes, while mice completely lacking RING1B exhibit gastrulation arrest.

REFERENCES

1. Lee, S.J., et al. 2001. E3 ligase activity of RING finger proteins that interact with Hip-2, a human ubiquitin-conjugating enzyme. FEBS Lett. 503: 61-64.
2. Suzuki, M., et al. 2002. Involvement of the Polycomb-group gene Ring1B in the specification of the anterior-posterior axis in mice. Development 129: 4171-4183.
3. Tuckfield, A., et al. 2002. Binding of the RING polycomb proteins to specific target genes in complex with the grainyhead-like family of developmental transcription factors. Mol. Cell. Biol. 22: 1936-1946.
4. Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 608985. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/
5. Voncken, J.W., et al. 2003. Rnf2 (Ring1b) deficiency causes gastrulation arrest and cell cycle inhibition. Proc. Natl. Acad. Sci. USA 100: 2468-2473.

CHROMOSOMAL LOCATION

Genetic locus: RNF2 (human) mapping to 1q25.3.

PRODUCT

RING1B 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 RING1B shRNA Plasmid (h): sc-62946-SH and RING1B shRNA (h) Lentiviral Particles: sc-62946-V as alternate gene silencing products.

For independent verification of RING1B (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-62946A, sc-62946B and sc-62946C.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.

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

RING1B siRNA (h) is recommended for the inhibition of RING1B 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

RING1B (N-32): sc-101109 is recommended as a control antibody for monitoring of RING1B 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 RING1B gene expression knockdown using RT-PCR Primer: RING1B (h)-PR: sc-62946-PR (20 µl, 523 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.