

RBP1 siRNA (h): sc-62930

BACKGROUND

RB (retinoblastoma protein) is a potent transcriptional regulator that is directly involved with events such as entry into cell division and formation of heterochromatin. RBP1 (retinoblastoma-binding protein 1), also known as RBP-1, RBBP1 or ARID4A (AT-rich interactive domain-containing protein 4A), is a ubiquitously expressed nuclear protein that binds directly to the viral-binding domain of RB. RB recruits chromatin-modifying proteins, such as RBP1, that can bind to it and allow it to act as a transcriptional repressor of E2F target genes. Once bound to RB, RBP1 can also act as a bridging molecule to recruit histone deacetylases (HDACs), proteins that function as potent regulators of gene expression. Three isoforms of RBP1 exist due to alternative splicing events.

REFERENCES

1. Lai, A., Kennedy, B.K., Barbie, D.A., Bertos, N.R., Yang, X.J., Theberge, M.C., Tsai, S.C., Seto, E., Zhang, Y., Kuzmichev, A., Lane, W.S., Reinberg, D., Harlow, E. and Branton, P.E. 2001. RBP1 recruits the mSIN3-histone deacetylase complex to the pocket of retinoblastoma tumor suppressor family proteins found in limited discrete regions of the nucleus at growth arrest. *Mol. Cell. Biol.* 21: 2918-2932.
2. Chen, Y.F., Chiu, H.H., Wu, C.H., Wang, J.Y., Chen, F.M., Tzou, W.H., Shin, S.J. and Lin, S.R. 2003. Retinoblastoma protein (pRB) was significantly phosphorylated through a Ras-to-MAPK pathway in mutant K-Ras stably transfected human adrenocortical cells. *DNA Cell Biol.* 22: 657-664.
3. Meehan, W.J., Samant, R.S., Hopper, J.E., Carrozza, M.J., Shevde, L.A., Workman, J.L., Eckert, K.A., Verderame, M.F. and Welch, D.R. 2004. Breast cancer metastasis suppressor 1 (BRMS1) forms complexes with retinoblastoma-binding protein 1 (RBP1) and the mSin3 histone deacetylase complex and represses transcription. *J. Biol. Chem.* 279: 1562-1569.
4. Binda, O., Roy, J.S. and Branton, P.E. 2006. RBP1 family proteins exhibit SUMOylation-dependent transcriptional repression and induce cell growth inhibition reminiscent of senescence. *Mol. Cell. Biol.* 26: 1917-1931.
5. Monroe, D.G., Secreto, F.J., Hawse, J.R., Subramaniam, M., Khosla, S. and Spelsberg, T.C. 2006. Estrogen receptor isoform-specific regulation of the retinoblastoma-binding protein 1 (RBBP1) gene: roles of AF1 and enhancer elements. *J. Biol. Chem.* 281: 28596-28604.

CHROMOSOMAL LOCATION

Genetic locus: ARID4A (human) mapping to 14q23.1.

PRODUCT

RBP1 siRNA (h) is a target-specific 19-25 nt siRNA designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see RBP1 shRNA Plasmid (h): sc-62930-SH and RBP1 shRNA (h) Lentiviral Particles: sc-62930-V as alternate gene silencing products.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

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

RBP1 siRNA (h) is recommended for the inhibition of RBP1 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 μ M in 66 μ l. 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

RBP1 (T-19): sc-55749 is recommended as a control antibody for monitoring of RBP1 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 donkey anti-goat IgG-HRP: sc-2020 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible donkey anti-goat IgG-HRP: sc-2033 (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 donkey anti-goat IgG-FITC: sc-2024 (dilution range: 1:100-1:400) or donkey anti-goat IgG-TR: sc-2783 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor RBP1 gene expression knockdown using RT-PCR Primer: RBP1 (h)-PR: sc-62930-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

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