

BAT1 siRNA (m): sc-62014

BACKGROUND

BAT1 (spliceosome RNA helicase BAT1), also known as DEAD-box protein UAP56 (56 kDa U2AF65-associated protein), HLA-B associated transcript-1 or ATP-dependent RNA helicase p47, is a member of the DECD subfamily of DEAD-box helicases. Important for mRNA splicing and nuclear export, BAT1 interacts with the mRNA export factor ALY and also functions as a splicing factor, mediating the first ATP-dependent step of spliceosome assembly. BAT1 associates with transcription elongation factor THO proteins, forming the TREX complex, and also interacts with splicing machinery to form the exon junction complex. Due to alternative splicing events, BAT1 exists in two isoforms. DDX39 (DEAD box protein 39), like BAT1, is a member of the DEAD-box family of helicases. Localized to the nucleus and expressed in lung, brain, kidney, spleen, thymus and salivary gland, DDX39 functions in a similar manner to BAT1 and is involved in pre-mRNA splicing and mRNA export out of the nucleus. DDX39 expression is upregulated in lung squamous cell carcinoma, suggesting a role for DDX39 in tumorigenesis.

REFERENCES

1. Fleckner, J., et al. 1997. U2AF65 recruits a novel human DEAD box protein required for the U2 snRNP-branchpoint interaction. *Genes Dev.* 11: 1864-1872.
2. Momose, F., et al. 2001. Cellular splicing factor RAF-2p48/NPI-5/BAT1/UAP56 interacts with the influenza virus nucleoprotein and enhances viral RNA synthesis. *J. Virol.* 75: 1899-1908.
3. Luo, M.L., et al. 2001. Pre-mRNA splicing and mRNA export linked by direct interactions between UAP56 and Aly. *Nature* 413: 644-647.
4. Price, P., et al. 2004. Polymorphisms at positions -22 and -348 in the promoter of the BAT1 gene affect transcription and the binding of nuclear factors. *Hum. Mol. Genet.* 13: 967-974.
5. Shi, H., et al. 2004. Crystal structure of the human ATP-dependent splicing and export factor UAP56. *Proc. Natl. Acad. Sci. USA* 101: 17628-17633.
6. Thakurta, A.G., et al. 2005. Homolog of BRCA2-interacting Dss1p and Uap56p link Mlo3p and Rae1p for mRNA export in fission yeast. *EMBO J.* 24: 2512-2523.

CHROMOSOMAL LOCATION

Genetic locus: Ddx39b (mouse) mapping to 17 B1.

PRODUCT

BAT1 siRNA (m) 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see BAT1 shRNA Plasmid (m): sc-62014-SH and BAT1 shRNA (m) Lentiviral Particles: sc-62014-V as alternate gene silencing products.

For independent verification of BAT1 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-62014A, sc-62014B and sc-62014C.

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

BAT1 siRNA (m) is recommended for the inhibition of BAT1 expression in mouse 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

BAT1/DDX39 (H-6): sc-271395 is recommended as a control antibody for monitoring of BAT1 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-IgG κ BP-HRP: sc-516102 or m-IgG κ 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-IgG κ BP-FITC: sc-516140 or m-IgG κ 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 BAT1 gene expression knockdown using RT-PCR Primer: BAT1 (m)-PR: sc-62014-PR (20 μ l). 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 for detailed protocols and support products.