

## YB-1 siRNA (r): sc-63323

### BACKGROUND

Y-box binding protein YB-1 (also known as CCAAT-binding transcription factor, enhancer factor I subunit A and DNA-binding protein B) belongs to a family of multifunctional proteins which regulate both transcription and translation. Y-box proteins interact with a wide variety of nucleic acid structures to act as transcription factors and mRNA masking proteins. The modular structure of Y-box proteins includes a highly conserved N-terminal cold-shock domain (CSD, equivalent to the bacterial cold-shock proteins) and four basic C-terminal domains containing Arginine clusters and aromatic residues. YB-1 plays a role in cell proliferation as an activator of growth-associated gene expression. YB-1 is also a repressor of the cell death-associated gene FAS. YB-1 may play an important role in controlling cell survival by regulating the expression of cell growth-associated and death-associated genes.

### REFERENCES

- Okamoto, T., et al. 2000. Direct interaction of p53 with the Y-box binding protein, YB-1: a mechanism for regulation of human gene expression. *Oncogene* 19: 6194-6202.
- Levenson, V.V., et al. 2000. Pleiotropic re-sistance to DNA-interactive drugs is associated with increased expression of genes involved in DNA replication, repair, and stress response. *Cancer Res.* 60: 5027-5030.
- Wang, N., et al. 2000. Acquisition of double-stranded DNA-binding ability in a hybrid protein between *Escherichia coli* CspA and the cold shock domain of human YB-1. *Mol. Microbiol.* 38: 526-534.
- Lasham, A., et al. 2000. Regulation of the human FAS promoter by YB-1, Pur $\alpha$  and AP-1 transcription factors. *Gene* 252: 1-13.

### CHROMOSOMAL LOCATION

Genetic locus: Ybx1 (rat) mapping to 5q36.

### PRODUCT

YB-1 siRNA (r) 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see YB-1 shRNA Plasmid (r): sc-63323-SH and YB-1 shRNA (r) Lentiviral Particles: sc-63323-V as alternate gene silencing products.

For independent verification of YB-1 (r) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-63323A, sc-63323B and sc-63323C.

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

### APPLICATIONS

YB-1 siRNA (r) is recommended for the inhibition of YB-1 expression in rat 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  $\mu$ M in 66  $\mu$ 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

YB-1 (C-3): sc-398340 is recommended as a control antibody for monitoring of YB-1 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 $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  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 $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  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 YB-1 gene expression knockdown using RT-PCR Primer: YB-1 (r)-PR: sc-63323-PR (20  $\mu$ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

### SELECT PRODUCT CITATIONS

- Shi, J.H., et al. 2012. Retinoic acid receptor  $\alpha$  mediates all-*trans*-retinoic acid-induced Klf4 gene expression by regulating Klf4 promoter activity in vascular smooth muscle cells. *J. Biol. Chem.* 287: 10799-10811.
- Shi, J.H., et al. 2013. Novel insight into Y-box binding protein 1 in the regulation of vascular smooth muscle cell proliferation through targeting GC box-dependent genes. *FEBS Lett.* 587: 1326-1332.

### RESEARCH USE

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

### PROTOCOLS

See our web site at [www.scbt.com](http://www.scbt.com) for detailed protocols and support products.