

# ASB-9 siRNA (h): sc-91095

## BACKGROUND

Members of the suppressor of cytokine signaling (SOCS) family of proteins contain C-terminal regions of homology called the SOCS box, which serves to couple SOCS proteins and their binding partners with the Elongin B and C complex. Several other families of proteins also contain SOCS boxes, but differ from the SOCS proteins in the type of domain they contain upstream of the SOCS box. The largest family of SOCS box-containing proteins is the ankyrin repeat and SOCS box-containing (ASB) protein family. Members of the ASB family include ASB-1 through ASB-18 and are involved in a variety of biological processes. ASB-9 is a 294 amino acid member of this family. It contains six ankyrin repeats and one SOCS box domain. ASB-9 functions as a ubiquitin ligase and, via its SOCS box domain, it specifically interacts with creatine kinase-B, targeting it for degradation and regulating its expression within the cell. Two isoforms exist for ASB-9 due to alternative splicing events.

## REFERENCES

1. Bork, P. 1993. Hundreds of ankyrin-like repeats in functionally diverse proteins: mobile modules that cross phyla horizontally? *Proteins* 17: 363-374.
2. Hilton, D.J., et al. 1998. Twenty proteins containing a C-terminal SOCS box form five structural classes. *Proc. Natl. Acad. Sci. USA* 95: 114-119.
3. Kile, B.T., et al. 2000. Cloning and characterization of the genes encoding the ankyrin repeat and SOCS box-containing proteins ASB-1, ASB-2, ASB-3 and ASB-4. *Gene* 258: 331-341.
4. Kile, B.T., et al. 2002. The SOCS box: a tale of destruction and degradation. *Trends Biochem. Sci.* 27: 235-241.
5. Kohroki, J., et al. 2005. ASB proteins interact with Cullin5 and Rbx2 to form E3 ubiquitin ligase complexes. *FEBS Lett.* 579: 6796-6802.
6. Debrincat, M.A., et al. 2007. Ankyrin repeat and suppressors of cytokine signaling box protein ASB-9 targets creatine kinase-B for degradation. *J. Biol. Chem.* 282: 4728-4737.

## CHROMOSOMAL LOCATION

Genetic locus: ASB9 (human) mapping to Xp22.2.

## PRODUCT

ASB-9 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see ASB-9 shRNA Plasmid (h): sc-91095-SH and ASB-9 shRNA (h) Lentiviral Particles: sc-91095-V as alternate gene silencing products.

For independent verification of ASB-9 (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-91095A, sc-91095B and sc-91095C.

## PROTOCOLS

See our web site at [www.scbt.com](http://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  $\mu$ 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

ASB-9 siRNA (h) is recommended for the inhibition of ASB-9 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  $\mu$ 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

ASB-9 (E-12): sc-166723 is recommended as a control antibody for monitoring of ASB-9 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 ASB-9 gene expression knockdown using RT-PCR Primer: ASB-9 (h)-PR: sc-91095-PR (20  $\mu$ 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.