

# Bag-5 siRNA (m): sc-72605

## BACKGROUND

Bag-5 (Bcl-2-associated athanogene 5), also known as Bag family molecular chaperone regulator 5, is a member of the Bag family of proteins and contains four Bag domains. Via their Bag domain, Bag proteins bind with high affinity to the HSC 70/HSP 70 ATPase domain, regulating chaperone activity and apoptosis. Bag-5 is a component of the HSP 70/Parkin complex and acts to inhibit Parkin E3 ubiquitin ligase activity and HSP 70 chaperone activity. In this complex, Bag-5 directly interacts with the ATPase domain of HSP 70 and the N-terminal linker region of Parkin. Bag-5 expression is induced upon dopaminergic neuron injury and functions to sensitize the neurons to injury-induced cell death. In addition, Bag-5 may be a useful target in therapies for neurodegenerative diseases such as Parkinson's disease which is caused by a mutation in the gene encoding Parkin.

## REFERENCES

1. Höhfeld, J., et al. 1997. GrpE-like regulation of the hsc70 chaperone by the anti-apoptotic protein BAG-1. *EMBO J.* 16: 6209-6216.
2. Takayama, S., et al. 1999. An evolutionarily conserved family of Hsp70/Hsc70 molecular chaperone regulators. *J. Biol. Chem.* 274: 781-786.
3. Brikanová, K., et al. 2002. BAG4/SODD protein contains a short BAG domain. *J. Biol. Chem.* 277: 31172-31178.
4. Kalia, S.K., et al. 2004. BAG5 inhibits parkin and enhances dopaminergic neuron degeneration. *Neuron.* 44: 931-945.
5. Esser, C., et al. 2004. Cooperation of molecular chaperones with the ubiquitin/proteasome system. *Biochim. Biophys. Acta.* 1695: 171-188.
6. Liman, J., et al. 2005. Interaction of BAG1 and Hsp70 mediates neuroprotectivity and increases chaperone activity. *Mol. Cell. Biol.* 25: 3715-3725.
7. Wada, S., et al. 2006. A genomewide analysis of genes for the heat shock protein 70 chaperone system in the ascidian *Ciona intestinalis*. *Cell Stress Chaperones.* 11: 23-33.

## CHROMOSOMAL LOCATION

Genetic locus: Bag5 (mouse) mapping to 12.

## PRODUCT

Bag-5 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Bag-5 shRNA Plasmid (m): sc-72605-SH and Bag-5 shRNA (m) Lentiviral Particles: sc-72605-V as alternate gene silencing products.

For independent verification of Bag-5 (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-72605A, sc-72605B and sc-72605C.

## PROTOCOLS

See our web site at [www.scbt.com](http://www.scbt.com) or our catalog 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

Bag-5 siRNA (m) is recommended for the inhibition of Bag-5 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  $\mu$ M in 60  $\mu$ 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

Bag-5 (D-20): sc-68957 is recommended as a control antibody for monitoring of Bag-5 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 Bag-5 gene expression knockdown using RT-PCR Primer: Bag-5 (m)-PR: sc-72605-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.