SANTA CRUZ BIOTECHNOLOGY, INC.

Bim siRNA (m): sc-29803

BACKGROUND

Pro-apototic Bcl-2 family members promote cell death by neutralizing their anti-apoptotic relatives, which otherwise maintain cell viability by regulating caspase activity. Bim belongs to the BH3-only subgroup of Bcl-2 related proteins and exists in three distinct isoforms, Bim_S (short), Bim_L (long) and Bim_{EL} (extra long). ERK1/2 phosphorylates Bim_{EL}, resulting in rapid degradation of the isoform via the proteasome pathway. At least three sites for ERK1/2 phosphorylation exist on Bim_{EL}, whereas ERK1/2 does not effect Bim_S or Bim_L, implying a unique role for Bim_{EL} in cell survival signaling.

REFERENCES

1. O'Reilly, L.A., et al. 2000. The proapoptotic BH3-only protein Bim is expressed in hematopoietic, epithelial, neuronal, and germ cells. Am. J. Pathol. 157: 449-461.
2. Seward, R.J., et al. 2003. Phosphorylation of the pro-apoptotic protein Bim in lymphocytes is associated with protection from apoptosis. Mol. Immunol. 39: 983-993.
3. Luciano, F., et al. 2003. Phosphorylation of Bim_{EL} by ERK1/2 on serine 69 promotes its degradation via the proteasome pathway and regulates its proapoptotic function. Oncogene 22: 6785-6793.
4. Ley, R., et al. 2004. Extracellular signal-regulated kinases 1/2 are serum-stimulated "Bim_{EL} kinases" that bind to the BH3-only protein Bim_{EL} causing its phosphorylation and turnover. J. Biol. Chem. 279: 8837-8847.
5. Harada, H., et al. 2004. Survival factor-induced extracellular signal-regulated kinase phosphorylates Bim, inhibiting its association with BAX and proapoptotic activity. Proc. Natl. Acad. Sci. USA 101: 15313-15317.

CHROMOSOMAL LOCATION

Genetic locus: Bcl2l11 (mouse) mapping to 2 F1.

PRODUCT

Bim 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Bim shRNA Plasmid (m): sc-29803-SH and Bim shRNA (m) Lentiviral Particles: sc-29803-V as alternate gene silencing products.

For independent verification of Bim (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-29803A, sc-29803B and sc-29803C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

Bim siRNA (m) is recommended for the inhibition of Bim 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 µM in 66 µl. 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

Bim (H-5): sc-374358 is recommended as a control antibody for monitoring of Bim 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 Bim gene expression knockdown using RT-PCR Primer: Bim (m)-PR: sc-29803-PR (20 µl, 590 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Liu, J., et al. 2008. Bcr-Abl mutants spread resistance to non-mutated cells through a paracrine mechanism. Leukemia 22: 791-799.
2. Murphy, B.M., et al. 2010. Contrasting patterns of Bim induction and neuroprotection in Bim-deficient mice between hippocampus and neocortex after status epilepticus. Cell Death Differ. 17: 459-468.
3. Qian, L., et al. 2011. miR-24 inhibits apoptosis and represses Bim in mouse cardiomyocytes. J. Exp. Med. 208: 549-560.
4. Hahm, E.R. and Singh, S.V. 2012. Bim contributes to phenethyl isothiocyanate-induced apoptosis in breast cancer cells. Mol. Carcinog. 51: 465-474.

RESEARCH USE

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