SANTA CRUZ BIOTECHNOLOGY, INC.

BLNK siRNA (m): sc-29811

BACKGROUND

Cross-linking of the B cell receptor (BCR) activates a variety of signaling pathways involved in processes such as cell proliferation and apoptosis. Intracellular protein tyrosine kinases such as Syk and Lyn have been implicated in this BCR signal transduction and are thought to play an important role in B cell development. BLNK (for B cell linker protein) is a central linker protein in B cells which has been shown to associate with the effector proteins GRB2, Vav, Nck and PLC γ following activation of the B cell receptor. The two forms of BLNK, pp68 and pp70, arise from alternate splicing of the human BLNK gene transcript. BLNK is phosphorylated by the Syk tyrosine kinase, which in turn permits activation of downstream effector proteins including GRB2 and PLC γ.

REFERENCES

1. DeFranco, A.L. 1997. The complexity of signaling pathways activated by the BCR. Curr. Opin. Immunol. 9: 296-308.
2. Kurosaki, T. 1997. Molecular mechanisms in B cell antigen receptor signaling. Curr. Opin. Immunol. 9: 309-318.
3. Reth, M., et al. 1997. Initiation and processing of signals from the B cell antigen receptor. Annu. Rev. Immunol. 15: 453-479.
4. Fu, C., et al. 1997. Identification of two tyrosine phosphoproteins, pp70 and pp68, which interact with phospholipase C γ, GRB2 and Vav after B cell antigen receptor activation. J. Biol. Chem. 272: 27362-27368.
5. Fu, C., et al. 1998. BLNK: a central linker protein in B cell activation. Immunity 9: 93-103.

CHROMOSOMAL LOCATION

Genetic locus: Blnk (mouse) mapping to 19 C3.

PRODUCT

BLNK siRNA (m) is a pool of 2 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 BLNK shRNA Plasmid (m): sc-29811-SH and BLNK shRNA (m) Lentiviral Particles: sc-29811-V as alternate gene silencing products.

For independent verification of BLNK (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-29811A and sc-29811B.

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

BLNK siRNA (m) is recommended for the inhibition of BLNK 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

BLNK (2B11): sc-8003 is recommended as a control antibody for monitoring of BLNK 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 BLNK gene expression knockdown using RT-PCR Primer: BLNK (m)-PR: sc-29811-PR (20 µl, 597 bp). 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.