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CIB siRNA (m): sc-43272

BACKGROUND

Platelets regulate the function of Integrin $\alpha 2b/\beta 3$ (GPIIb/IIIa), the platelet Fibrinogen receptor, which is involved in the binding of proteins to integrin cytoplasmic domains. A novel protein, CIB, for calcium- and integrin-binding protein (also designated as Kip for kinase interacting protein, SIP2-28 and DNA-PK_{CS} interacting protein), binds specifically at the cytoplasmic domain of $\alpha 2b$ by a number of positively charged residues in its binding site. Binding of CIB to the $\alpha 2b$ is affected by fluctuations in the intracellular calcium concentration. In aggregated platelets, endogenous CIB and $\alpha 2b/\beta 3$ translocate to the Triton X-100-insoluble cytoskeleton, demonstrating that the cellular localization of CIB is regulated. CIB also binds to DNA-PK_{CS}, which is a nuclear protein serine/threonine kinase that plays a role in the DNA repair and recombination process of lymphoid development. Fnk also binds to the CIB, suggesting that CIB may be a regulatory subunit of polo-like kinases. CIB shows significant homology to calcineurin B and calmodulin, and its mRNA levels are ubiquitously expressed in various human tissues.

REFERENCES

- Naik, U.P., et al. 1997. Identification of a novel calcium-binding protein that interacts with the Integrin $\alpha 2b$ cytoplasmic domain. *J. Biol. Chem.* 272: 4651-4654.
- Wu, X. and Lieber, M.R. 1997. Interaction between DNA-dependent protein kinase and a novel protein, Kip. *Mutat. Res.* 385: 13-20.
- Shock, D.D., et al. 1999. Calcium-dependent properties of CIB binding to the Integrin $\alpha 2b$ cytoplasmic domain and translocation to the platelet cytoskeleton. *Biochem. J.* 342: 729-735.
- Seki, N., et al. 1999. Structure, expression profile and chromosomal location of an isolog of DNA-PK_{CS} interacting protein (Kip) gene. *Biochim. Biophys. Acta* 1444: 143-147.
- Hwang, P.M. and Vogel, H.J. 2000. Structures of the platelet calcium- and integrin-binding protein and the Integrin $\alpha 2b$ cytoplasmic domain suggest a mechanism for calcium-regulated recognition; homology modeling and NMR studies. *J. Mol. Recognit.* 13: 83-92.

CHROMOSOMAL LOCATION

Genetic locus: Cib1 (mouse) mapping to 7 D3.

PRODUCT

CIB 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see CIB shRNA Plasmid (m): sc-43272-SH and CIB shRNA (m) Lentiviral Particles: sc-43272-V as alternate gene silencing products.

For independent verification of CIB (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-43272A, sc-43272B and sc-43272C.

RESEARCH USE

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

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

APPLICATIONS

CIB siRNA (m) is recommended for the inhibition of CIB 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 μ M in 66 μ 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

CIB (G-5): sc-271490 is recommended as a control antibody for monitoring of CIB 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 κ BP-HRP: sc-516102 or m-IgG κ 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 κ BP-FITC: sc-516140 or m-IgG κ 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 CIB gene expression knockdown using RT-PCR Primer: CIB (m)-PR: sc-43272-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.