

PBP siRNA (h): sc-39363

BACKGROUND

Members of the α -chemokine subfamily of inducible, secreted, pro-inflammatory cytokines contain a similar motif, in which the first two cysteine residues are separated by a single residue (Cys-X-Cys), and are also chemotactic for neutrophils. The platelet basic protein (PBP), a member of the α -chemokine family, resides in the α -granules of platelets and is released upon their activation. Proteolytic cleavage of the amino terminus of PBP leads to the generation of several peptides, which include mature PBP, connective tissue-activating peptide III (CTAP III, also designated low affinity platelet factor IV (LA-PF4)), β -thromboglobulin (β -TG), and neutrophil-activating peptide 2 (NAP-2). PBP and its N-truncated derivatives mediate inflammation and wound healing. Specifically, NAP-2 activates chemotaxis and degranulation in neutrophils during inflammation. The gene encoding human PBP maps to chromosome 4q13.3.

REFERENCES

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- Wenger, R.H., et al. 1991. Human platelet basic protein/connective tissue activating peptide-III maps in a gene cluster on chromosome 4q12-q13 along with other genes of the β -thromboglobulin superfamily. *Hum. Genet.* 87: 367-368.
- Car, B.D., et al. 1991. Formation of neutrophil-activating peptide 2 from platelet-derived connective-tissue-activating peptide III by different tissue proteinases. *Biochem. J.* 275: 581-584.
- Hoogewerf, A.J., et al. 1995. CXC chemokines connective tissue activating peptide-III and neutrophil activating peptide-2 are heparin/heparan sulfate-degrading enzymes. *J. Biol. Chem.* 270: 3268-3277.
- Malkowski, M.G., et al. 1997. The amino-terminal residues in the crystal structure of connective tissue activating peptide-III (des10) block the ELR chemotactic sequence. *J. Mol. Biol.* 266: 367-380.
- Proudfoot, A.E., et al. 1997. Structure and bioactivity of recombinant human CTAP-III and NAP-2. *J. Protein Chem.* 16: 37-49.

CHROMOSOMAL LOCATION

Genetic locus: PPBP (human) mapping to 4q13.3.

PRODUCT

PBP 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see PBP shRNA Plasmid (h): sc-39363-SH and PBP shRNA (h) Lentiviral Particles: sc-39363-V as alternate gene silencing products.

For independent verification of PBP (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-39363A, sc-39363B and sc-39363C.

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

PBP siRNA (h) is recommended for the inhibition of PBP 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 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

PBP (C-24): sc-73636 is recommended as a control antibody for monitoring of PBP 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 PBP gene expression knockdown using RT-PCR Primer: PBP (h)-PR: sc-39363-PR (20 μ 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.

PROTOCOLS

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