SANTA CRUZ BIOTECHNOLOGY, INC.

Crossveinless-2 siRNA (m): sc-72319

BACKGROUND

Crossveinless-2, also known as BMP-binding endothelial regulator protein, Cvl2 or Cv2, is a member of the Chordin family. It is an evolutionarily conserved protein that was first identifed in *Drosophila* where it is required for the formation of cross-veins in the wing. Crossveinless-2 is a developmentally secreted glycoprotein that contains a trypsin inhibitory-like (TIL) domain, five von Willebrand factor type C (VWFC) domains and one VWF type D (VWFD) domain. Crossveinless-2 regulates BMP homeostasis in early vertebrate embryonic tissues via its cysteine-rich BMP-binding domains. It is expressed during development at sites of high BMP signaling and its expression is responsive to this signaling thereby providing positive feedback. Crossveinless-2 directly interacts with BMP4 and BMP2 and can function either to enhance or inhibit BMP signaling. Crossveinless-2 may function to promote BMP signaling by aiding in ligand transport.

REFERENCES

1. Coffinier, C., et al. 2003. Mouse Crossveinless-2 is the vertebrate homolog of a *Drosophila* extracellular regulator of BMP signaling. Gene Expr. Patterns 2: 189-194.
2. Coles, E., et al. 2004. A vertebrate crossveinless 2 homologue modulates BMP activity and neural crest cell migration. Development 131: 5309-5317.
3. Binnerts, M.E., et al. 2004. Human Crossveinless-2 is a novel inhibitor of bone morphogenetic proteins. Biochem. Biophys. Res. Commun. 315: 272-280.
4. Kamimura, M., et al. 2004. Vertebrate crossveinless 2 is secreted and acts as an extracellular modulator of the BMP signaling cascade. Dev. Dyn. 230: 434-445.
5. O'Connor, M.B., et al. 2005. Shaping BMP morphogen gradients in the *Drosophila* embryo and pupal wing. Development 133: 183-193.
6. Ikeya, M., et al. 2006. Essential pro-Bmp roles of crossveinless 2 in mouse organogenesis. Development 133: 4463-4473.

CHROMOSOMAL LOCATION

Genetic locus: Bmper (mouse) mapping to 9 A3.

PRODUCT

Crossveinless-2 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 Crossveinless-2 shRNA Plasmid (m): sc-72319-SH and Crossveinless-2 shRNA (m) Lentiviral Particles: sc-72319-V as alternate gene silencing products.

For independent verification of Crossveinless-2 (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-72319A, sc-72319B and sc-72319C.

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 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

Crossveinless-2 siRNA (m) is recommended for the inhibition of Crossveinless-2 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

Crossveinless-2 (G-8): sc-377502 is recommended as a control antibody for monitoring of Crossveinless-2 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 Crossveinless-2 gene expression knockdown using RT-PCR Primer: Crossveinless-2 (m)-PR: sc-72319-PR (20 µl). 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.