

# SCUBE2 siRNA (h): sc-96423

## BACKGROUND

SCUBE2 (signal peptide, CUB domain, EGF-like 2), also known as CEGP1, Cegb1 or Cegf1, is a 999 amino acid protein that is ubiquitously expressed in adult tissues and belongs to the evolutionarily conserved SCUBE protein family. Containing a CUB domain and nine EGF-like domains, SCUBE2 manifests as a secreted surface-anchored glycoprotein when overexpressed and is considered a novel component of the HH (Hedgehog) signal. The HH signal plays a pivotal role in induction of ventral neuronal and muscle cell types around the midline during vertebrate development. It is suggested that SCUBE2 expression is important in breast cancer progression and may serve as a useful prognostic marker. SCUBE2 forms homo-oligomers and hetero-oligomers with SCUBE1 and SCUBE3. Expressed as three isoforms produced by alternative splicing events, SCUBE2 is encoded by a gene located on human chromosome 11, which houses over 1,400 genes and comprises nearly 4% of the human genome.

## REFERENCES

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3. Kawakami, A., et al. 2005. The zebrafish-secreted matrix protein you/SCUBE2 is implicated in long-range regulation of Hedgehog signaling. *Curr. Biol.* 15: 480-488.
4. Woods, I.G. and Talbot, W.S. 2005. The you gene encodes an EGF-CUB protein essential for Hedgehog signaling in zebrafish. *PLoS Biol.* 3: e66.
5. Hollway, G.E., et al. 2006. SCUBE2 mediates Hedgehog signalling in the zebrafish embryo. *Dev. Biol.* 294: 104-118.
6. Tsai, M.T., et al. 2009. Isolation and characterization of a secreted, cell-surface glycoprotein SCUBE2 from humans. *Biochem. J.* 422: 119-128.
7. Cheng, C.J., et al. 2009. SCUBE2 suppresses breast tumor cell proliferation and confers a favorable prognosis in invasive breast cancer. *Cancer Res.* 69: 3634-3641.

## CHROMOSOMAL LOCATION

Genetic locus: SCUBE2 (human) mapping to 11p15.4.

## PRODUCT

SCUBE2 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see SCUBE2 shRNA Plasmid (h): sc-96423-SH and SCUBE2 shRNA (h) Lentiviral Particles: sc-96423-V as alternate gene silencing products.

For independent verification of SCUBE2 (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-96423A, sc-96423B and sc-96423C.

## APPLICATIONS

SCUBE2 siRNA (h) is recommended for the inhibition of SCUBE2 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  $\mu$ M in 66  $\mu$ 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

SCUBE2 (G-4): sc-398607 is recommended as a control antibody for monitoring of SCUBE2 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker<sup>™</sup> compatible goat anti-mouse IgG-HRP: sc-2031 (dilution range: 1:2000-1:5000), Cruz Marker<sup>™</sup> Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz<sup>™</sup> Mounting Medium: sc-24941.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor SCUBE2 gene expression knockdown using RT-PCR Primer: SCUBE2 (h)-PR: sc-96423-PR (20  $\mu$ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

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

## RESEARCH USE

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