### SANTA CRUZ BIOTECHNOLOGY, INC.

# INSIG-1 (H-70): sc-98985



BACKGROUND

INSIG-1 and INSIG-2 play distinct roles in a negative-feedback mechanism for cholesterol synthesis. INSIG-1 is highly expressed in liver and fibroblast cell lines. INSIG-1 localizes to the endoplasmic reticulum (ER) and binds the sterol-sensing domain of SREBP cleavage-activating protein (SCAP). Sterol induces INSIG-1 binding to SCAP. INSIG-2, another ER protein, binds SCAP in a sterol-regulated manner. Thus, INSIG-1 and INSIG-2 block the export of SCAP from the ER and ultimately inhibit cholesterol synthesis by preventing the proteolytic processing of SREBPs by Golgi enzymes. INSIG-1 is encoded by the Insulin-induced gene (INSIG-1). INSIG-1 gene expression is suppressed by oxysterols and restored following the introduction of the hypocholesterol-emic agent LY295427. The negative feedback mechanism is absent in mutant CHO cells with a point mutation in one SCAP allele within the sterol-sensing domain. The mutant cells constitutively cleave SREBP in the presence of sterols. The critical role of INSIG-1 and INSIG-2 in cholesterol metabolism may be exploited as a therapeutic effect for hypercholesterolemia.

#### REFERENCES

- Peng, Y., Schwarz, E.J., Lazar, M.A., Genin, A., Spinner, N.B. and Taub, R. 1997. Cloning, human chromosomal assignment, and adipose and hepatic expression of the CL-6/INSIG1 gene. Genomics 43: 278-284.
- Janowski, B.A. 2002. The hypocholesterolemic agent LY295427 upregulates INSIG-1, identifying the INSIG-1 protein as a mediator of cholesterol homeostasis through SREBP. Proc. Natl. Acad. Sci. USA 99: 12675-12680.
- Yabe, D., Brown, M.S. and Goldstein, J.L. 2002. Insig-2, a second endoplasmic reticulum protein that binds SCAP and blocks export of sterol regulatory element-binding proteins. Proc. Natl. Acad. Sci. USA 99: 12753-12758.
- Yang, T., Espenshade, P.J., Wright, M.E., Yabe, D., Gong, Y., Aebersold, R., Goldstein, J.L. and Brown, M.S. 2002. Crucial step in cholesterol homeostasis: sterols promote binding of SCAP to INSIG-1, a membrane protein that facilitates retention of SREBPs in ER. Cell 110: 489-500.
- Yabe, D., Xia, Z.P., Adams, C.M. and Rawson, R.B. 2002. Three mutations in sterol-sensing domain of SCAP block interaction with INSIG and render SREBP cleavage insensitive to sterols. Proc. Natl. Acad. Sci. USA 99: 16672-16677.

#### CHROMOSOMAL LOCATION

Genetic locus: INSIG1 (human) mapping to 7q36.2.

#### SOURCE

INSIG-1 (H-70) is a rabbit polyclonal antibody raised against amino acids 31-100 mapping near the N-terminus of INSIG-1 of human origin.

#### PRODUCT

Each vial contains 200  $\mu g$  lgG in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

#### **RESEARCH USE**

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

#### APPLICATIONS

INSIG-1 (H-70) is recommended for detection of INSIG-1 of human origin by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000), immunoprecipitation [1-2  $\mu$ g per 100-500  $\mu$ g of total protein (1 ml of cell lysate)], immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500) and solid phase ELISA (starting dilution 1:30, dilution range 1:30-1:3000).

Suitable for use as control antibody for INSIG-1 siRNA (h): sc-44432, INSIG-1 shRNA Plasmid (h): sc-44432-SH and INSIG-1 shRNA (h) Lentiviral Particles: sc-44432-V.

Molecular Weight of INSIG-1: 30 kDa.

#### **RECOMMENDED SECONDARY REAGENTS**

To ensure optimal results, the following support (secondary) reagents are recommended: 1) Western Blotting: use goat anti-rabbit IgG-HRP: sc-2004 (dilution range: 1:2000-1:100,000) or Cruz Marker<sup>™</sup> compatible goat anti-rabbit IgG-HRP: sc-2030 (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) Immunoprecipitation: use Protein A/G PLUS-Agarose: sc-2003 (0.5 ml agarose/2.0 ml). 3) Immunofluorescence: use goat anti-rabbit IgG-FITC: sc-2012 (dilution range: 1:100-1:400) or goat anti-rabbit IgG-TR: sc-2780 (dilution range: 1:100-1:400) with UltraCruz<sup>™</sup> Mounting Medium: sc-24941.

#### **STORAGE**

Store at 4° C, \*\*D0 NOT FREEZE\*\*. Stable for one year from the date of shipment. Non-hazardous. No MSDS required.

#### PROTOCOLS

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

## MONOS

Satisfation Guaranteed

Try **INSIG-1 (A-9): sc-390504**, our highly recommended monoclonal alternative to INSIG-1 (H-70).