

GRASP65 (G-9): sc-365412

BACKGROUND

The Golgi apparatus is a highly complex organelle comprised of a stack of cisternal membranes on the secretory pathway from the ER to the cell surface. The structure is maintained by an exoskeleton or Golgi matrix constructed from a family of coiled-coil protein, the golgins and other peripheral membrane components such as GRASP55 and GRASP65. GRASP55 (Golgi reassembly stacking protein or p59) is a component of the Golgi stacking machinery. GRASP55 is highly homologous to GRASP65 and contains two PDZ domains. GRASP55 is myristoylated and palmitoylated. Unlike GRASP65, GRASP55 does not have detectable binding with the vesicle docking protein GM130 and is located on the medial-Golgi rather than *cis*-Golgi. Both GRASP55 and GRASP65 function in the stacking of Golgi cisternae. The novel coiled-coil protein golgin 45 interacts with GRASP55 and the GTP form of Rab 2, suggesting that GRASP55 and golgin 45 form a Rab 2 effector complex on medial-Golgi essential for normal protein transport and Golgi structure. ERK2 directly phosphorylates GRASP55, which is phosphorylated in mitotic cells, suggesting that mitogen-activated protein kinase (MKK)/ERK pathway phosphorylates the Golgi during mitosis.

REFERENCES

- Shorter, J., Watson, R., Giannakou, M.E., Clarke, M., Warren, G. and Barr, F.A. 1999. GRASP55, a second mammalian GRASP protein involved in the stacking of Golgi cisternae in a cell-free system. *EMBO J.* 18: 4949-4960.
- Kuo, A., Zhong, C., Lane, W.S. and Derynck, R. 2000. Transmembrane transforming growth factor α tethers to the PDZ domain-containing, Golgi membrane-associated protein p59/GRASP55. *EMBO J.* 19: 6427-6439.
- Short, B., Preisinger, C., Korner, R., Kopajtich, R., Byron, O. and Barr, F.A. 2001. A GRASP55-Rab 2 effector complex linking Golgi structure to membrane traffic. *J. Cell Biol.* 155: 877-883.
- Barr, F.A., Preisinger, C., Kopajtich, R. and Korner, R. 2001. Golgi matrix proteins interact with p24 cargo receptors and aid their efficient retention in the Golgi apparatus. *J. Cell Biol.* 155: 885-891.
- Jesch, S.A., Lewis, T.S., Ahn, N.G. and Linstedt, A.D. 2001. Mitotic phosphorylation of Golgi reassembly stacking protein 55 by mitogen-activated protein kinase ERK2. *Mol. Biol. Cell* 12: 1811-1817.

CHROMOSOMAL LOCATION

Genetic locus: GORASP1 (human) mapping to 3p22.2.

SOURCE

GRASP65 (G-9) is a mouse monoclonal antibody raised against amino acids 211-440 mapping at the C-terminus of GRASP65 of human origin.

PRODUCT

Each vial contains 200 μ g IgG_{2b} kappa light chain 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

GRASP65 (G-9) is recommended for detection of GRASP65 of human origin by Western Blotting (starting dilution 1:100, dilution range 1:100-1:1000), immunoprecipitation [1-2 μ g per 100-500 μ g of total protein (1 ml of cell lysate)], immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500), immunohistochemistry (including paraffin-embedded sections) (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 GRASP65 siRNA (h): sc-41228, GRASP65 shRNA Plasmid (h): sc-41228-SH and GRASP65 shRNA (h) Lentiviral Particles: sc-41228-V.

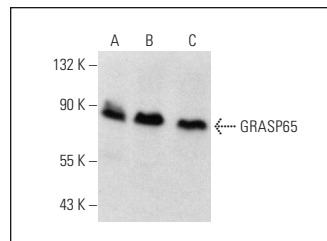
Molecular Weight of GRASP65: 65 kDa.

Positive Controls: MCF7 whole cell lysate: sc-2206, HeLa whole cell lysate: sc-2200 or HT-29 whole cell lysate: sc-364232.

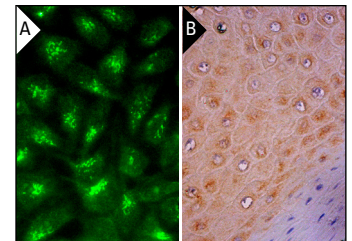
RECOMMENDED SUPPORT REAGENTS

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) Immunoprecipitation: use Protein A/G PLUS-Agarose: sc-2003 (0.5 ml agarose/2.0 ml). 3) 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. 4) Immunohistochemistry: use m-IgG κ BP-HRP: sc-516102 with DAB, 50X: sc-24982 and Immunohistomount: sc-45086, or Organo/Limonene Mount: sc-45087.

DATA



GRASP65 (G-9): sc-365412. Western blot analysis of GRASP65 expression in HeLa (A), MCF7 (B) and HT-29 (C) whole cell lysates.



GRASP65 (G-9): sc-365412. Immunofluorescence staining of methanol-fixed HeLa cells showing nuclear and cytoplasmic localization (A). Immunoperoxidase staining of formalin fixed, paraffin-embedded human oral mucosa tissue showing cytoplasmic staining of squamous epithelial cells (B).

STORAGE

Store at 4° C, **DO 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 for detailed protocols and support products.