

BACKGROUND

Ras p21 is the prototype of a superfamily of GTPases that is involved in the regulation of a wide variety of cellular processes. Ras signals in its GTP-bound form but is "turned off" when bound to GDP. When unregulated or constitutively turned on by mutations, Ras signaling contributes to malignant transformation. The switch between active and inactive Ras is controlled by GTPase-activating proteins (GAPs) and guanine nucleotide exchange factors (GEFs). C3G was isolated in a screen for proteins that could bind the SH3 domain of the Crk proto-oncogene product. The carboxy-terminus of the C3G protein displays significant sequence similarity to Ras-GRF/Cdc25Mm and mSos and can substitute for Cdc25 function in *S. cerevisiae*. These observations strongly suggest that C3G is a GEF for Ras and is involved in the regulation of Ras signaling through Crk. The C3G gene maps to human chromosome 9q34.13 in proximity to the gene that encodes c-Abl, a proto-oncogene that regulates Crk.

REFERENCES

- Mochizuki, N., et al. 2000. Crk activation of JNK via C3G and R-Ras. *J. Biol. Chem.* 275: 12667-12671.
- Ohba, Y., et al. 2001. Requirement for C3G-dependent Rap1 activation for cell adhesion and embryogenesis. *EMBO J.* 20: 3333-3341.
- Zhai, B., et al. 2001. C3G, a guanine nucleotide exchange factor bound to adapter molecule c-Crk, has two alternative splicing forms. *Biochem. Biophys. Res. Commun.* 286: 61-66.
- Voss, A.K., et al. 2003. The guanine nucleotide exchange factor C3G is necessary for the formation of focal adhesions and vascular maturation. *Development* 130: 355-367.
- Ling, L., et al. 2003. Src-CrkII-C3G-dependent activation of Rap1 switches growth hormone-stimulated p44/42 MAP kinase and JNK/SAPK activities. *J. Biol. Chem.* 278: 27301-27311.

CHROMOSOMAL LOCATION

Genetic locus: Rargef1 (mouse) mapping to 2 B.

SOURCE

p-C3G (G-2) is a mouse monoclonal antibody specific for a short amino acid sequence containing Tyr 514 phosphorylated C3G of mouse origin.

PRODUCT

Each vial contains 200 µg IgM kappa light chain in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

Blocking peptide available for competition studies, sc-365994 P, (100 µg peptide in 0.5 ml PBS containing < 0.1% sodium azide and 0.2% stabilizer protein).

STORAGE

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

APPLICATIONS

p-C3G (G-2) is recommended for detection of Tyr 514 phosphorylated C3G of mouse origin by Western Blotting (starting dilution 1:100, dilution range 1:100-1:1000), immunoprecipitation [1-2 µg per 100-500 µg 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).

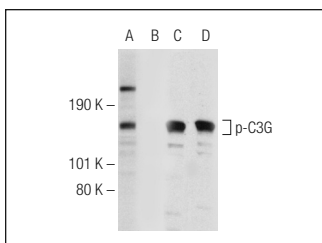
Suitable for use as control antibody for C3G siRNA (m): sc-29864, C3G shRNA Plasmid (m): sc-29864-SH and C3G shRNA (m) Lentiviral Particles: sc-29864-V.

Molecular Weight of p-C3G: 135 kDa.

Positive Controls: K-562 whole cell lysate: sc-2203.

RECOMMENDED SUPPORT REAGENTS

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, TBS Blotto B Blocking Reagent: sc-2335 (use 50 mM NaF, sc-24988, as diluent), Lambda Phosphatase: sc-200312A and Western Blotting Luminol Reagent: sc-2048. 2) Immunoprecipitation: use Protein L-Agarose: sc-2336 (0.5 ml agarose/2.0 ml). 3) 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.

DATA

Western blot analysis of C3G phosphorylation in untreated (A,C) and lambda protein phosphatase (sc-200312A) treated (B,D) K-562 whole cell lysates. Antibodies tested include p-C3G (G-2): sc-365994 (A,B) and C3G (H-300): sc-15359 (C,D).

SELECT PRODUCT CITATIONS

- Dar, M.I., et al. 2020. Differentiation of human neuroblastoma cell line IMR-32 by sildenafil and its newly discovered analogue IS00384. *Cell. Signal.* 65: 109425.
- Ishii, K., et al. 2022. Reelin regulates the migration of late-born hippocampal CA1 neurons via cofilin phosphorylation. *Mol. Cell. Neurosci.* 124: 103794.

RESEARCH USE

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