Datasheet for ABIN968482

anti-PIK3C2B antibody (AA 16-209)

2 Images | 1 Publication

Go to Product page

Overview

Quantity:	50 µg
Target:	PIK3C2B
Binding Specificity:	AA 16-209
Reactivity:	Human
Host:	Mouse
Clonality:	Monoclonal
Conjugate:	This PIK3C2B antibody is un-conjugated
Application:	Western Blotting (WB), BioImaging (BI)

Product Details

Immunogen:	Human PI3-Kinase C2beta aa. 16-209
Clone:	22-PI3
Isotype:	IgG1
Characteristics:	1. Since applications vary, each investigator should titrate the reagent to obtain optimal results. 2. Please refer to us for technical protocols. 3. This antibody has been developed and certified for the bioimaging application. However, a routine bioimaging test is not performed on every lot. Researchers are encouraged to titrate the reagent for optimal performance. 4. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.

Product Details

	5. Source of all serum proteins is from USDA inspected abattoirs located in the United States. 6. Triton is a trademark of the Dow Chemical Company.
Purification:	The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

Target Details

Target:	PIK3C2B
Alternative Name:	PI3-Kinase C2 beta (PIK3C2B Products)
Background:	Phosphatidylinositol (PtdIns) (3) kinase phosphorylates the D-3 position of the inositolring of PtdIns, producing PtdIns(3)P, PtdIns(3,4)P2, and PtdIns(3,4,5)P3. PI3-kinase is a heterodimer of an 85 kDa regulatory subunit (p85) and a 110 kDa catalytic subunit (p110). However, it is only one member of a larger family of proteins with similarity to the p110 subunit. These different PI3-kinase isoforms have been divided into three classes. Class I consists of p110alpha and p110ß which bind the p85 subunit and associate with receptor tyrosine kinases. Class II includes 68D and cpk from Drosophila, p170 and cpk-m from mouse, and C2alpha, C2ß (HsC2), and C2gamma from human. These proteins phosphorylate PtdIns and PtdIns(4)P, but not PtdIns(4,5)P2, and each contain a C-terminal C2 domain that may negatively regulate the catalytic domain. Class III members only phosphorylate PtdIns to PtdIns(3)P and include the S. cerevisiae Vps34p and its human homologs. In humans, the class II PI3-kinases C2alpha and C2ß have similar catalytic, PI kinase, and C2 domains. However they differ in their N-terminal regions. In addition, C2ß has no cation specificity, while C2alpha prefers Mg2+-ATP for optimal phosphorylation.
Molecular Weight:	165 kDa
Pathways:	Inositol Metabolic Process

Application Details

Application Notes:	Bioimaging 1. Seed the cells in appropriate culture medium at ~10,000 cells per well in an 96-well Imaging Plate and culture overnight. 2. Remove the culture medium from the wells, and fix the cells by adding 100 myl of Fixation Buffer to each well. Incubate for 10 minutes at room temperature (RT). 3. Remove the fixative from the wells, and permeabilize the cells using either 90% methanol, or Triton™ X-100: a. Add 100 myl of -20°C 90% methanol to each well and incubate for 5 minutes

Application Details

	at RT. OR b. Add 100 myl of 0.1% Triton™ X-100 to each well and incubate for 5 minutes at RT. 4. Remove the permeabilization buffer, and wash the wells twice with 100 myl of 1× PBS. 5. Remove the PBS, and block the cells by adding 100 myl of to each well. Incubate for 30 minutes at RT. 6. Remove the blocking buffer and add 50 myl of the optimally titrated primary antibody (diluted in Stain Buffer) to each well, and incubate for 1 hour at RT. 7. Remove the primary antibody, and wash the wells three times with 100 myl of 1× PBS. 8. Remove the PBS, and add the second step reagent at its optimally titrated concentration in 50 myl to each well, and incubate in the dark for 1 hour at RT. 9. Remove the second step reagent, and wash the wells three times with 100 myl of 1× PBS. 10. Remove the PBS, and counter-stain the nuclei by adding 200 myl per well of 2 myg/ml Hoechst 33342 in 1× PBS to each well at least 15 minutes before imaging. 11. View and analyze the cells on an appropriate imaging instrument.
Comment:	Related Products: ABIN967389, ABIN968535
Restrictions:	For Research Use only

Handling

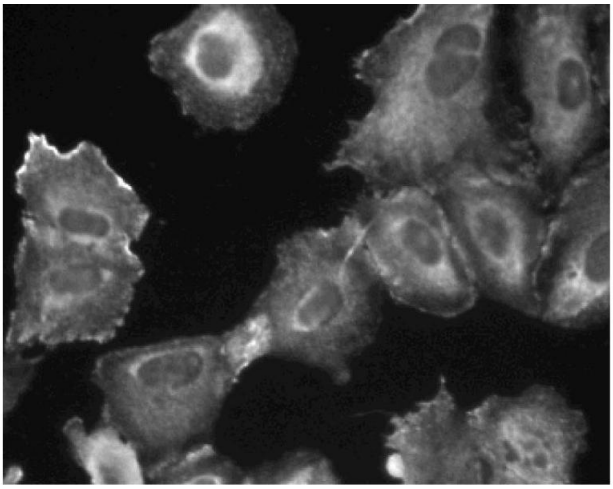
Format:	Liquid
Concentration:	250 µg/mL
Buffer:	Aqueous buffered solution containing BSA, glycerol, and ≤0.09 % sodium azide.
Preservative:	Sodium azide
Precaution of Use:	This product contains Sodium azide: a POISONOUS AND HAZARDOUS SUBSTANCE which should be handled by trained staff only.
Storage:	-20 °C
Storage Comment:	Store undiluted at -20°C.

Publications

Product cited in:	Arcaro, Volinia, Zvelebil, Stein, Watton, Layton, Gout, Ahmadi, Downward, Waterfield: "Human phosphoinositide 3-kinase C2beta, the role of calcium and the C2 domain in enzyme activity." in: **The Journal of biological chemistry**, Vol. 273, Issue 49, pp. 33082-90, (1999) (PubMed).

Western Blotting

Image 1. Western blot analysis of PI3-Kinase C2beta on a HeLa lysate. Lane 1: 1:1000, lane 2: 1:2000, lane 3: 1:4000 dilution of the PI3-Kinase C2beta antibody.



Immunofluorescence

Image 2. Immunofluorescent staining of A549 (ATCC CCL-185) cells. Cells were seeded in a 96 well imaging plate at ~ 10 000 cells per well. After overnight incubation, cells were stained using the alcohol perm protocol and the anti-PI3-Kinase C2beta antibody. The second step reagent was FITC goat anti mouse Ig. The image was taken on a BD Pathway 855 Bioimager using a 20x objective. This antibody also stained U-2 OS (ATCC HTB-96) and HeLa (ATCC CCL-2) cells using both the Triton™ X-100 and alcohol perm protocols.