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# Datasheet for ABIN967708 anti-PIK3CA antibody (AA 562-724)

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#### Overview

Quantity:	150 µg
Target:	PIK3CA
Binding Specificity:	AA 562-724
Reactivity:	Human, Mouse, Rat, Dog, Chicken
Host:	Mouse
Clonality:	Monoclonal
Conjugate:	This PIK3CA antibody is un-conjugated
Application:	Western Blotting (WB), Immunohistochemistry (IHC), Immunoprecipitation (IP), BioImaging (BI)

## Product Details

Immunogen:	Human PI3-Kinase alpha subunit aa. 562-724
Clone:	4-PI3
lsotype:	lgG2a
Cross-Reactivity:	Dog (Canine), Rat (Rattus), Mouse (Murine), Chicken
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

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### Target Details

Target:	PIK3CA
Alternative Name:	PI3-Kinase (PIK3CA Products)
Background:	PI3-kinase phosphorylates the D-3 position of the inositol ring of phosphatidylinositol (PtdIns),
	PtdIns(4)P and PtdIns(4,5)P2 to produce the respective PI3-phosphorylated derivatives. PI3-
	kinase exists as a heterodimer of 85 kDa (p85) and 110 kDa (p110) subunits. The p85 subunit
	contains two SH2 domains and an SH3 domain. It associates with and serves as a substrate
	for activated growth factor receptor tyrosine kinases. p85 may serve as regulator of the
	catalytic subunit, p110, by acting as the link between PI3-kinase and the ligand-activated
	receptor. Two distinct forms of the p85 subunit have been described: 1) p85alpha, which binds
	tightly to the catalytic subunit, and 2) p85ß, a protein whose function is presently unknown.
	Both isoforms bind to activated receptors and serve as tyrosine kinase substrates.
Molecular Weight:	85 kDa
Pathways:	PI3K-Akt Signaling, RTK Signaling, TCR Signaling, AMPK Signaling, Interferon-gamma Pathway,
	TLR Signaling, Fc-epsilon Receptor Signaling Pathway, EGFR Signaling Pathway, Neurotrophin
	Signaling Pathway, Inositol Metabolic Process, Hepatitis C, CXCR4-mediated Signaling Events,
	Signaling Events mediated by VEGFR1 and VEGFR2, Signaling of Hepatocyte Growth Factor
	Receptor, VEGFR1 Specific Signals, VEGF Signaling

### Application Details

Application Notes:	Bioimaging
	1. Seed the cells in appropriate culture medium at $\sim$ 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

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	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 $1 \times PBS$ .
	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 $1 \times PBS$ .
	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 $1 \times PBS$ .
	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, ABIN968533
Restrictions:	For Research Use only

### Handling

Format:	Liquid
Concentration:	250 µg/mL
Buffer:	Aqueous buffered solution containing BSA, glycerol, and $\leq 0.09$ % 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:Efendiev, Yudowski, Zwiller, Leibiger, Katz, Berggren, Pedemonte, Leibiger, Bertorello: "Relevance of dopamine signals anchoring dynamin-2 to the plasma membrane during Na+,K+-ATPase endocytosis." in: The Journal of biological chemistry, Vol. 277, Issue 46, pp. 44108-14,(2002) (PubMed).

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Nguyen, Ho, Beattie, Barber: "TEL-JAK2 mediates constitutive activation of the phosphatidylinositol 3'-kinase/protein kinase B signaling pathway." in: **The Journal of biological chemistry**, Vol. 276, Issue 35, pp. 32704-13, (2001) (PubMed).

Zhang, Bontrager, Hemler: "Transmembrane-4 superfamily proteins associate with activated protein kinase C (PKC) and link PKC to specific beta(1) integrins." in: **The Journal of biological chemistry**, Vol. 276, Issue 27, pp. 25005-13, (2001) (PubMed).

Cantley, Auger, Carpenter, Duckworth, Graziani, Kapeller, Soltoff: "Oncogenes and signal transduction." in: **Cell**, Vol. 64, Issue 2, pp. 281-302, (1991) (PubMed).

#### Images



#### Western Blotting

**Image 1.** Western blot analysis of PI3-Kinase on a A431 lysate. Lane 1: 1:2500, lane 2: 1:5000, lane 3: 1:10000 dilution of the PI3-Kinase antibody.

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#### 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 antibody. The second step reagent was FITC goat anti mouse Ig. The image was taken on a BD Pathway<sup>™</sup> 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<sup>™</sup> X-100 and alcohol perm protocols.

#### Image 3.

