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Technical Data Sheet

For research use only
Not intended or approved for
diagnostic or therapeutic use.

PI3K α E545K – Active Mutant (Human p110 α /p85 α)

Full-length recombinant protein expressed in Sf9 cells

Catalog Number:	P27-15H
Size:	10 μ g
Concentration:	0.1 mg/mL, in 50 mM sodium phosphate, pH 7.0, 300 mM NaCl, 150 mM imidazole, 0.1 mM PMSF, 0.25 mM DTT, and 25% glycerol.
Description:	Recombinant full-length human p110 α mutant (E545K) and human p85 α co-expressed by baculovirus in Sf9 insect cells using an N-terminal His tag on both proteins. The gene accession number is NM_006218 for p110 α , and NM_181523 for p85 α .
Gene Aliases:	p110 α : PIK3CA, p110-alpha, MGC142163 p85 α : PIK3R1, GRB1, p85-alpha
Specificity:	Converts PI(4,5)P ₂ to PI(3,4,5)P ₃ <i>in vivo</i> ; converts PI to PI(3)P, and PI(4)P to PI(3,4)P ₂ <i>in vitro</i> .
Assay Condition:	50 mM Hepes, pH 7.4, 50 mM NaCl, 5 mM MgCl ₂ , 0.05% CHAPS, 2 mM DTT, 25 μ M ATP, containing 5 μ M PI(4,5)P ₂ substrate and 0.05 – 0.5 ng/ μ L PI3K α (optimize by enzyme titration as needed). Perform reactions at room temperature for 30 - 90 min.
Suggested Use:	PI3K activity assay and PI3K α mutant (E545K) inhibitor screening.
Storage:	Store product at -70 °C or below. Enzyme is stable for 6 months from date of shipment. For best result, aliquot enzyme into small quantities after centrifugation, flash freeze aliquots, and store at recommended temperature. Avoid repeated handling and multiple freeze/thaw cycles.
Background:	Phosphatidylinositol 3-kinases (PI3Ks) are heterodimers that function in cellular pathways downstream of receptor tyrosine kinase (RTK) and G-protein coupled receptors (GPCRs). PI3Ks are central players in normal cell homeostasis as well as cancer, heart disease, and inflammatory disorders. Class I PI3K α contains a 110 kDa catalytic subunit (PIK3CA) that is frequently mutated and hyperactivated in up to 30% of common cancers including breast, colon, endometrium, and prostate. ^{1,2} The mutation of an aspartate residue at position 545 to lysine is quite common (hot spot mutation) and causes transformation. ³ The complete oncogenic mechanism and its clinical significance, however, remains to be elucidated. ⁴
References:	<ol style="list-style-type: none">1. Samuels, Y. et al. High frequency of mutations of the PIK3CA gene in human cancers. <i>Science</i> 304: 554 only, 2004.2. Kang, S. et al. Oncogenic transformation induced by the p110beta, -gamma, and -delta isoforms of class I phosphoinositide 3-kinase. <i>Proc Natl Acad Sci USA</i> 2006, 103, 1289-94.3. Bader, A. G. et al. Cancer-specific mutations in PIK3CA are oncogenic <i>in vivo</i>. <i>Proc Natl Acad Sci U S A</i> 2006, 103, 1475-9.4. Huang, C. H. et al. The structure of a human p110alpha/p85alpha complex elucidates the effects of oncogenic PI3Kalpha mutations. <i>Science</i> 2007, 318, 1744-8.
Related Products:	PI3K Activity Assay kits: K-1000S (ELISA), K-1100 (FP), and K-1300 (AlphaScreen™) Other PI3K enzymes: E-2000 (hPI3K α), P27-18H (mPI3K α), and P28-10H (hPI3K β). PI(4,5)P ₂ substrates: P-4508, C-45B6, C-45F6, C-45M6, H-45BT, H-45FL, H-45TR, H-45TM, and P-4516