

Recombinant GRP-PH Domain

CATALOG NUMBER: STA-200

STORAGE: -80°C; avoid freeze/thaw

QUANTITY AND CONCENTRATION: 100 µg at 1.0 mg/mL in 1X PBS

SHELF LIFE: 1 year from date of receipt under proper storage conditions

Background

Cellular 3-phosphoinositides are generated through the actions of a family of PI 3-kinases and appear to have regulatory roles in multiple cell functions, such as membrane ruffling, chemotaxis, secretory responses, insulin-mediated membrane translocation of glucose transporters, membrane trafficking of growth factor receptors, and regulated cell adhesion. In mammalian cells three classes of PI 3-kinases have been identified. These include p110 isoforms regulated by p85 subunits containing SRC homology 2 (SH2) domains, a p110 γ PI 3-kinase regulated by heterotrimeric guanine nucleotide-binding proteins, and a PI 3-kinase containing a homology domain (C2 domain) thought to bind membrane lipids. The p85-p110 and p110 γ type PI 3-kinases that are specifically activated by receptor signaling systems exhibit broad substrate specificities, and their activation leads to rapid phosphorylation of the inositol D-3 positions on PtdIns, PtdIns-4-phosphate [PtdIns(4)P], and PtdIns-4,5-bisphosphate [PtdIns(4,5)P₂].

Several protein targets of 3-phosphoinositides have been recently identified. These include certain protein kinase C isoforms and the pleckstrin homology (PH) domain-containing protein kinases c-Akt and Btk. GRP1 (general receptor for phosphoinositide) binds phosphatidylinositol-3,4,5-trisphosphate [PtdIns(3,4,5)P₃] through a pleckstrin homology (PH) domain and displays a region of high sequence similarity to the yeast Sec7 protein.

Purity and Activity

Expressed and purified from E.Coli as a fusion protein, greater than 90% by SDS-PAGE (Figure 1). The recombinant protein of GRP-PH domain selectively binds to PIP3 (Figure 2).

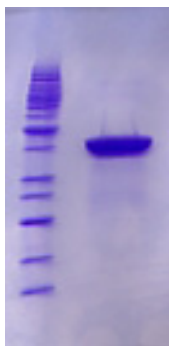


Figure 1. Recombinant GRP-PH on SDS-PAGE

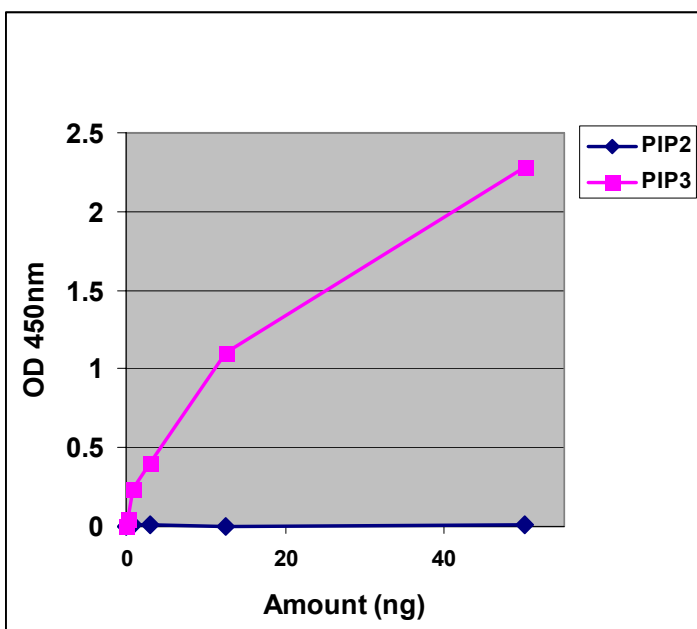


Figure 2. PIP3 Binding to GRP-PH. Biotinylated PI(3,4,5)P3 or PI(4,5)P2 is added to GRP-PH coated 96-well plate. After 1 hr incubation, the plate was washed and blocked with 1XPBS/1% BSA. The capture phosphoinositol was detected with Streptavidin-HRP and TMB substrate.

References

1. Klarlund JK, Guilherme A, Holik JJ, Virbasius JV, Chawla A, Czech MP. (1997) *Science* **275**, 1927-30.

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