

## Recombinant PRKACA protein

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**Catalog No:** 81179, 81879

**Lot No:** 16218001

**Expressed In:** Baculovirus

**Quantity:** 20, 1000 µg

**Concentration:** 0.15 µg/µl

**Source:** Human

**Buffer Contents:** Recombinant PRKACA protein is supplied in 25 mM HEPES-NaOH pH 7.5, 300 mM NaCl, 10% glycerol, 0.04% Triton X-100, 0.5 mM TCEP.

**Background:** PRKACA (Protein Kinase CAMP-Activated Catalytic Subunit Alpha), also called as PKACA or PPNAD4, is a member of the catalytic subunits of protein kinase A, which exists as a tetrameric holoenzyme with two regulatory subunits and two catalytic subunits, in its inactive form. cAMP causes the dissociation of the inactive holoenzyme into a dimer of regulatory subunits bound to four cAMP and two free monomeric catalytic subunits. Four different regulatory subunits and three catalytic subunits have been identified in humans. cAMP-dependent phosphorylation of proteins by protein kinase A is important to many cellular processes, including differentiation, proliferation, and apoptosis.

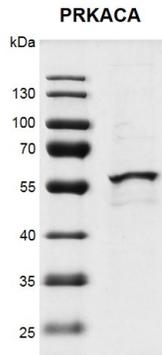
PRKACA can phosphorylate a large number of substrates in the cytoplasm and the nucleus. It regulates the abundance of compartmentalized pools of its regulatory subunits through phosphorylation of PJA2 which binds and ubiquitinates these subunits, leading to their subsequent proteolysis. PRKACA can phosphorylates CDC25B, ABL1, NFKB1, CLDN3, PSMC5/RPT6, PJA2, RYR2, RORA and VASP. It is involved in many important cellular processes such as glucose-mediated adipogenic differentiation and osteogenic differentiation, the regulation of platelets, embryonic development, and meiosis resumption.

**Protein Details:** Recombinant PRKACA protein was expressed in a baculovirus expression system as the full length protein (accession number NP\_002721.1) with an N-terminal FLAG tag. The molecular weight of the protein is 50.4 kDa.

**Application Notes:** Recombinant PRKACA protein is suitable for use in the study of enzyme kinetics, inhibitor screening, and selectivity profiling.

**Kinase Activity Assay Conditions:** 1 µM STK S2 substrate was incubated with different concentrations of PRKACA protein in a 10 µl reaction system containing 1×Enzymatic Buffer, 5 mM MgCl<sub>2</sub>, 1 mM DTT, 5 nM SEB and 100 µM ATP for 1 hr. Detection reagents containing STK antibody and SA-XL665, each of which was 1:100 diluted with 1×Detection Buffer, were added and incubated with the reactions for 30 min. All the operations and reactions were performed at room temperature, and HTRF KinEASE STK assay was used to detect the enzymatic activity.

**Storage and Guarantee:** Recombinant proteins in solution are temperature sensitive and must be stored at -80°C to prevent degradation. Avoid repeated freeze/thaw cycles and keep on ice when not in storage. This product is for research use only and is not for use in diagnostic procedures. This product is guaranteed for 6 months from date of arrival.



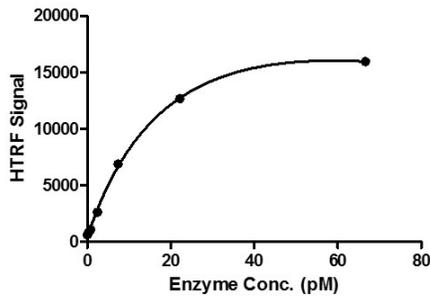
**Recombinant PRKACA protein**

10% SDS-PAGE Coomassie staining

MW: 50.4 kDa

Purity: ≥80%

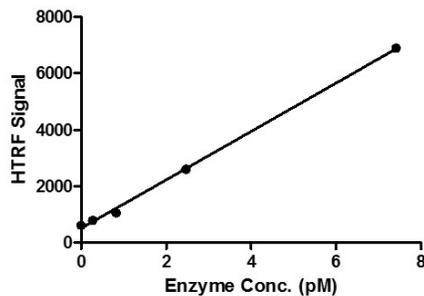
**PRKACA Titration**



**HTRF assay for recombinant PRKACA protein activity**

1  $\mu$ M STK S2 substrate was incubated with different concentrations of PRKACA protein in a 10  $\mu$ l reaction system for 1 hr. The 10  $\mu$ l detection reagents were added and incubated with the reactions for 30 min. All operations and reactions were performed at room temperature, and HTRF KinEASE STK assay was used to detect enzymatic activity.

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