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## Data Sheet

### PDE4A1A

Human, recombinant, N-terminal GST tag  
Catalog #: 60040

Formulated in: 25 mM Tris-HCl, pH 8.0,  
100 mM NaCl, 0.05% Tween-20, 50% glycerol,  
and 3 mM DTT.

Stability: >6 months at -80°C

References:

1. Sullivan, M., et al., Journal Biochem. J. 333:693-703 (1998).
2. Huston E, et al., J Cell Sci. 2006 119:3799-810. (2006).

Description:

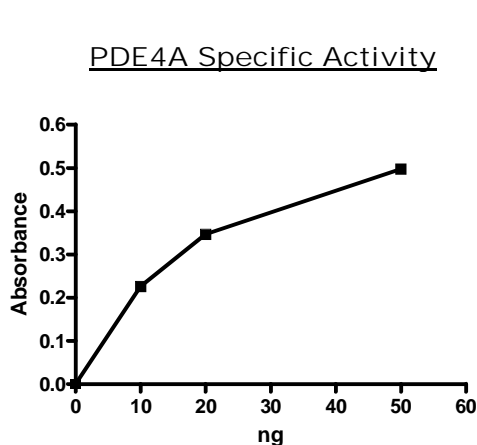
Human PDE4A1A (GenBank Accession No. U97584), full length with N-terminal GST tag, MW= 100 kDa, expressed in a Baculovirus infected Sf9 cell expression system.

Specific Activity: 4,176 pmol/min/μg.  
Assay conditions: 10 mM Tris-HCl, pH7.4,  
10 mM MgCl<sub>2</sub>, 1 mM MnCl<sub>2</sub>, 200 μM cAMP, 2.5 kU 5' nucleotidase, 37°C, 20min.

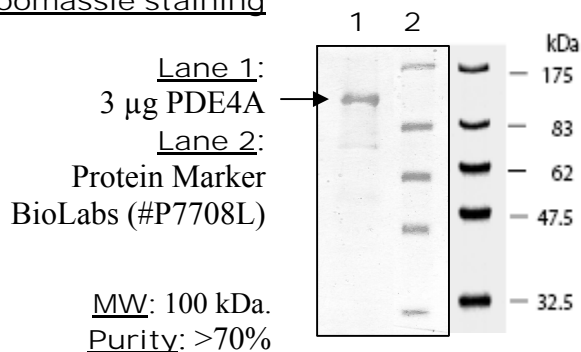
Application:

Useful for the study of enzyme kinetics, screening inhibitors, and selectivity profiling.

### Quality Assurance



10% SDS-PAGE  
Coomassie staining



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## Assay Protocol

**Materials:** IMAP™ TR-FRET Screening Express with Progressive Binding Kit from Molecular Devices (R8160); 2'-Fluo-AHC-cAMP from Axxora LLC (BLG-F003-01, San Diego, CA); PDE4A1A (BPS Catalog Number 60040).

### **Methods:**

#### **Step 1:**

Dilute FAM-cAMP to 200nM in 1X PDE Assay Buffer.

Dilute PDE4A1A enzymes to 2ng/μl in 1X PDE Assay Buffer.

#### **Step 2:**

Adding following components to a low binding black plate:

25μl of 200nM FAM-cAMP (Final concentration will be 100nM)

5μl of 1 x PDE assay buffer

20μl of PDE4A1A (2ng/μl) (Final amount=40ng/reaction)

Mix and incubate at room temperature for 1 hour.

#### **Step 3:**

Prepare 1X reagent-binding buffer (85% 1X Binding Buffer A and 15% 1X Binding Buffer B).

Prepare Binding Solution by diluting Binding Reagent with 1X reagent-binding Buffer (1:600).

Add 120μl of Binding Solution to each well and incubate the plate at room temperature for 1 hour.

#### **Step 4:**

Measure fluorescence polarization at excitation of 485nm and emission of 520nm in BioTek Synergy™ 2 microplate reader.

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