

TAMRA-cAMP PDE IV substrate *Red Fluorescence*

Catalog number: 13603

Unit size: 0.5 umol

Component	Storage	Amount
TAMRA-cAMP PDE IV substrate *Red Fluorescence*	Freeze (<-15 °C), Minimize light exposure	0.5 umol

OVERVIEW

This red cAMP derivative is a specific substrate for phosphodiesterase (PDE) IV. It can be used for assaying PDE IV activities or screening PDE IV inhibitors in combination with anti-cAMP antibody in a FRET readout or FP format. PDE is a group of enzymes that degrade the second messenger molecules: cyclic nucleotides cAMP and cGMP. They regulate the localization, duration, and amplitude of cyclic nucleotide signaling within subcellular domains. PDEs are therefore important regulators of signal transduction mediated by these second messenger molecules. PDE enzymes are often targets for pharmacological inhibition due to their unique tissue distribution, structural and functional properties. Inhibitors of PDE can prolong or enhance the effects of physiological processes mediated by cAMP or cGMP by inhibition of their degradation by PDE. PDE inhibitors have been identified as new potential therapeutics in areas such as pulmonary arterial hypertension, coronary heart disease, dementia, depression and schizophrenia.

AT A GLANCE

Important Following protocol only provides a guideline, and should be modified according to your specific needs.

KEY PARAMETERS

Instrument:	Fluorescence microplate reader
Excitation:	540 nm
Emission:	590 nm
Cutoff:	570 nm
Recommended plate:	Solid black

PREPARATION OF STOCK SOLUTIONS

Unless otherwise noted, all unused stock solutions should be divided into single-use aliquots and stored at -20 °C after preparation. Avoid repeated freeze-thaw cycles.

1. TAMRA-cAMP PDE IV stock solution (1 mM):

Make a 1 mM stock solution by adding 500 µL of DMSO into the vial of 0.5 umol TAMRA-cAMP PDE IV substrate.

Note Unused stock solution can be stored at -20 °C in dark place in single aliquotes.

PREPARATION OF WORKING SOLUTION

TAMRA-Cyclic-3', 5'-AMP PDE IV substrate assay solution (2X):

Make 2X TAMRA-Cyclic-3', 5'-AMP PDE IV substrate assay solution by diluting 1 mM TAMRA-Cyclic-3', 5'-AMP PDE IV substrate stock solution into your PDE buffer (such as 10 mM Tris-HCl, pH 7.4, 10 mM Mg Cl₂, 1 mM MnCl₂) to make a 200 - 400 nM solution.

Note Make only sufficient quantity needed for the assay.

SAMPLE EXPERIMENTAL PROTOCOL

- Mix equal volume of the PDE IV standards or samples with 2X TAMRA-Cyclic-3', 5'-AMP PDE IV substrate assay solution, and incubate at room temperature for at least 1 hour.
- Monitor the fluorescence polarization at Ex/Em = 540/590 nm.

EXAMPLE DATA ANALYSIS AND FIGURES

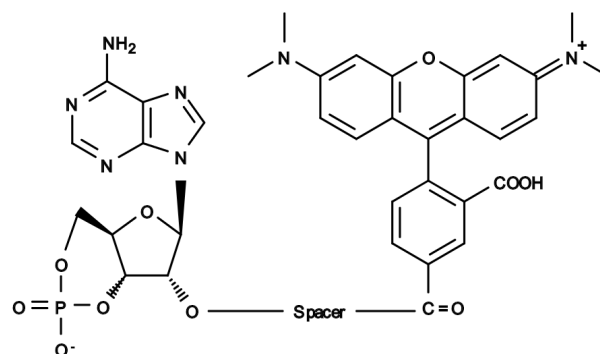


Figure 1. Chemical structure for TAMRA-cAMP PDE IV substrate *Red Fluorescence*

DISCLAIMER

AAT Bioquest provides high-quality reagents and materials for research use only. For proper handling of potentially hazardous chemicals, please consult the Safety Data Sheet (SDS) provided for the product. Chemical analysis and/or reverse engineering of any kit or its components is strictly prohibited without written permission from AAT Bioquest. Please call 408-733-1055 or email info@aatbio.com if you have any questions.