

### cAMP Assay Kit (Competitive ELISA) ab65355

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#### Overview

Product name	cAMP Assay Kit (Competitive ELISA)
Detection method	Colorimetric
Sample type	Urine, Serum, Other biological fluids, Tissue Extracts, Cell Lysate, Cell culture media
Assay type	Quantitative
Sensitivity	> 0.002 $\mu$ M
Range	0.002 $\mu$ M - 0.2 $\mu$ M
Assay time	3h 30m
Species reactivity	<b>Reacts with:</b> Mammals, Other species
Product overview	cAMP Assay Kit ab65355 uses a sensitive colorimetric competitive ELISA method to quantify cAMP (cyclic AMP) levels in samples from mammals and other species. It has a sensitivity of 0.002 $\mu$ M.

The cAMP assay protocol uses a 96-well plate supplied coated with Protein G. The Protein G binds an anti-cAMP antibody which is added to the plate.

HRP-labeled cAMP, and free cAMP within a sample, are then added to each well of the plate. The cAMP assay is based on the competition between the HRP-labeled cAMP and the free cAMP for the fixed number of cAMP antibody binding sites on the plate.

The more free cAMP is present, the less HRP-labeled cAMP is bound. After a washing step, the amount of bound HRP-labeled cAMP is detected using a colorimetric HRP substrate measured by absorbance at OD 450 nm. The level of signal is compared to a standard curve for free cAMP, which is run at the same time as the samples.

This kit uses an acetylation step to achieve the maximum sensitivity.

cAMP assay protocol summary:

- add prepared samples and standards to tubes
- add neutralizing buffer
- add acetylating reagent mix, vortex and incubate for 10 min at room temp
- add assay buffer
- transfer to protein-G 96-well plate

- add cAMP antibody and incubate for 1 hr at room temp
- add cAMP-HRP and incubate for 1 hr at room temp
- wash with assay buffer
- add HRP developer and incubate for 1 hr at room temp
- stop reaction with HCl
- analyze with microplate reader

**Notes** This product is manufactured by BioVision, an Abcam company and was previously called K371 cAMP Direct Immunoassay Kit (Colorimetric). K371-100 is the same size as the 100 test size of ab65355.

If you are using fluorometric detection, we recommend **cAMP Assay Kit (Fluorometric) (ab138880)**.

**Platform** Microplate reader

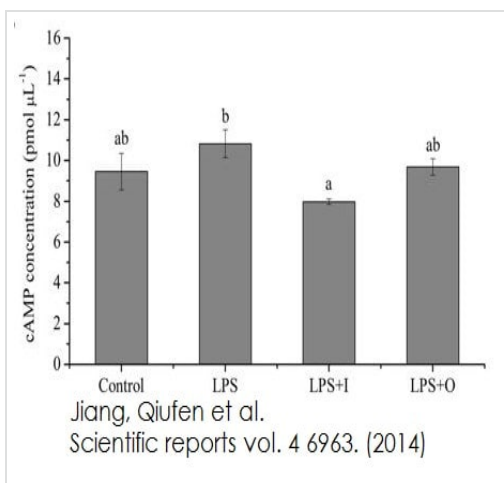
## Properties

**Storage instructions** Store at -20°C. Please refer to protocols.

Components	100 tests
Acetylating Reagent A	1 x 750µl
Acetylating Reagent B	1 x 1.5ml
Assay Buffer XXXVI	1 x 25ml
cAMP Standard	1 x 10µl
cAMP-HRP	1 vial
Neutralization Buffer III	1 x 7.5ml
Protein G Coated Plate	1 unit
Rabbit Anti-cAMP Ab	1 vial
TMB Substrate I	1 x 10ml

**Relevance** Adenosine 3',5'-cyclic monophosphate (cyclic AMP, cAMP) is an important "second messenger" involved in many physiological processes. It plays a key role as an intracellular second messenger for transduction events that follow a number of extracellular signals.

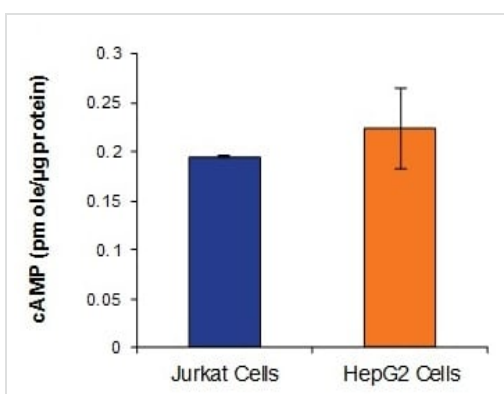
## Images



Functional Studies - cAMP Assay Kit (Competitive ELISA) (ab65355)

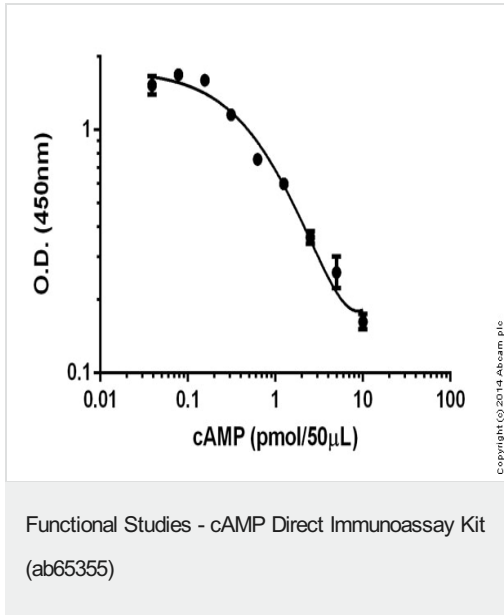
Jiang, Qiufen et al., Scientific reports vol. 4 6963., Fig 4, doi:10.1038/srep06963

Concentration of cAMP in response to in vitro haemocyte stimulation with LPS and  $\beta$ -AR (B) agonist/antagonist at 24h. Data presented as mean  $\pm$  S.D. (N = 6) of cAMP concentration expressed as pmol  $\mu\text{L}^{-1}$ . The significant differences among the control and treated groups were subjected to one-way ANOVA, followed by Student-Newman-Keuls multiple range test, which revealed two groups (a to b). The concentration of cAMP with the same letter in common do not differ at the P = 0.05 level of significance.



Functional Studies - cAMP Direct Immunoassay Kit (ab65355)

Measurement of cAMP in Jurkat and HepG2 cells with ab65355. Assay performed using the kit protocol.



**Please note:** All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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