

Assay

- Take 2 mL cells, shake at 200 rpm, at time 0, add 20 μ L 100 μ M cAMP.
- At time 0, 4, 8, 15, 30, 45, 60, 75, 90, 105, 120, 180 seconds take 100 μ L aliquots and add to 1mL assay buffer.
- Cover with aluminum foil and shake to fix at room temperature for 1 hr.
- Spin 14K for 10 min, keep pellet.
- Add 1 mL MeOH, wrap in aluminum foil and shake overnight.
- Read samples on Fluoro-Max. Excitation 540 nm, emission 570 nm, slitwidth 3mm.

Catalog # of Chemicals from Sigma

Rhodamine-Phalloidin	P-1951
Caffeine	C-0750
cAMP sodium salt	A-6885 (1 g)