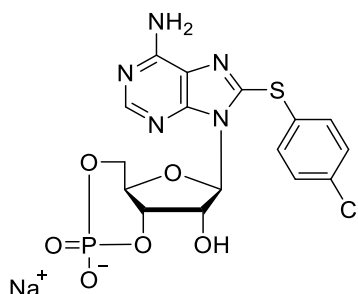


Technical Information about 8-(4-chlorophenylthio)-cAMP

Potent membrane-permeant activator of both cAMP- and cGMP-dependent protein kinases and of Epac

Update: July 03, 2018 HU



Abbreviation:

8-CPT-cAMP

Formula	CAS No.	Molecular Weight	UV	BIOLOG Cat.No.
C ₁₆ H ₁₄ ClN ₅ O ₆ PS·Na	[93882-12-3]	493.8	λ _{max} 282 nm / ε 16000 / pH 7	C 010

Name: 8- (4- Chlorophenylthio)adenosine- 3', 5'- cyclic monophosphate (8-CPT-cAMP) or *para*- chlorophenylthioadenosine- 3', 5'- cyclic monophosphate (8-pCPT-cAMP)

Description: 8-CPT-cAMP is an analogue of the natural signal molecule cyclic AMP in which the hydrogen in position 8 of the heterocyclic nucleobase is replaced by the lipophilic 4-chlorophenylthio moiety.

Properties: 8-CPT-cAMP is a potent stimulator of cAMP-dependent protein kinases and of the exchange protein activated by cyclic AMP (Epac). Due to its high lipophilicity (Braumann & Jastorff 1985) (> dibutyryl-cAMP), allowing good membrane permeability in most biosystems, and its good activation properties, 8-CPT-cAMP is very often used in signal transduction studies. However, good results obtained with 8-CPT-cAMP are not necessarily a definite proof of cAMP participation and we recommend being rather critical with published data and results concerning this analogue:

Caution: In contrast to its name, 8-CPT-cAMP is a potent stimulator of both, cAMP-and cGMP-dependent protein kinases (Miller et al. 1973, OGREID et al. 1989, Sandberg et al. 1991, Sugita et al. 1994)!

In addition, it is a rather good inhibitor of cGMP-specific PDE (V) and thus increases basal cGMP. Both properties result in lacking cA/cG pathway specificity (Connolly et al. 1992).

The compound is metabolized more slowly by PDE compared to cAMP, but is not as resistant as often stated, releasing metabolites (Coulson et al. 1983) with disturbing effects (Sandnes et al. 1996).

If a much more specific activator of protein kinase A is needed, we recommend 6-Phe-cAMP (Cat. No. P 006) or Sp-5,6-DCI-cBIMPS (Cat. No. D 014).

Interestingly, for specific activation of the cGMP pathway the corresponding 8-pCPT-cGMP (Cat. No. C 009) is a good choice.

BIOLOG also offers the metabolically stable phosphorothioates, the inhibitory Rp-8-CPT-cAMPS (Cat. No. C 011), and the agonistic Sp-8-CPT-cAMPS (Cat. No. C 012) (Singh et al. 1998), as well as potential metabolites of 8-CPT-cAMP such as 8-pCPT-5'-AMP (Cat. No. C 101) and 8-pCPT-Ado (Cat. No. C 086).

Specification: Lyophilized or crystallized sodium salt. The free acid or other salt forms are available upon request. Equal concentrations of 8-CPT-cAMP can appear very different in volume due to sensitivity of the lyophilized form to humidity. The compound can even contract to small volume droplets. Normallyly the product is located in the conical bottom of the tube. Micromolar quantities are determined by UV at λ_{max}.

Purity: Typical analysis is better than 99% (HPLC / UV / 282 nm). Traces of fluorescent impurities inevitably formed during production have been removed by an additional purification step. The product is not sterile and has not been tested for endotoxins.

Stability and Storage: 8-CPT-cAMP is chemically stable under conditions of biological systems and media. Nevertheless, we recommend that the compound should be stored in the freezer, for longer storage periods preferably in freeze-dried form.

Since UV radiation develops a fluorescent impurity which can disturb in fluorescence assays, avoid bright light during handling.

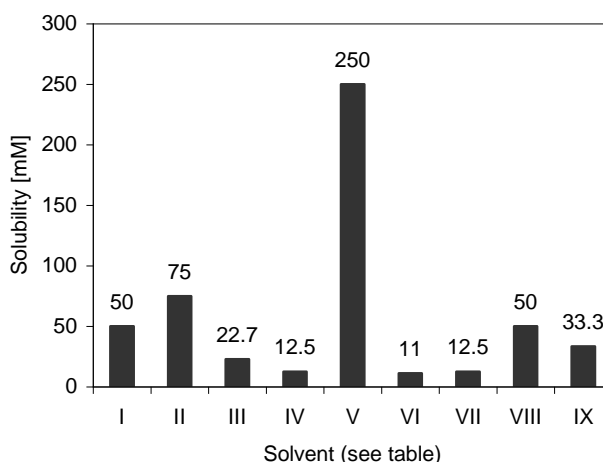
Toxicity and Safety: Since cyclic AMP has multiple tasks in every organism it is very likely that lipophilic cAMP analogues will interfere with many cell regulation processes *in vivo*. However, due to the rather small quantities to work with no health hazards have been reported. Nevertheless please keep in mind that the *in vivo* properties of this compound are not sufficiently characterized up to now. Avoid skin contact or ingestion and allow only trained personnel to handle the product.

Our products are designed, developed and sold for research purposes only. They are intended for *in vitro* and nonhuman *in vivo* laboratory applications. Any other use requires approval of health authorities.

Not for drug, household or related uses!

Solubility: Detailed information on the solubility of 8-pCPT-cAMP in water and various buffers are listed in the solubility chart below. Concentrations have been determined at ambient temperature and can be considered as minimum concentrations usually obtainable, however, slight batch-to-batch variations cannot be ruled out. When opening the tube please make sure that no substance is lost within the cap. Please rinse tube walls carefully and preferably use ultrasonic or vortex to achieve total and uniform mixing.

No.	Solvent	Solubility [mM]
I	H ₂ O	50
II	DMSO	75
III	DMF	22.7
IV	Ethanol 96%	12.5
V	Methanol	250
VI	PBS, pH 7.4	11
VII	100 mM Na ₂ HPO ₄ , pH 7.0	12.5
VIII	25 mM HEPES/NaOH, pH 7.2	50
IX	25 mM Tris/HCl, pH 7.4	33.3



Selected References for 8-CPT-cAMP:

Since 8-CPT-cAMP is a well known biochemical tool there exist numerous references for almost every biosystem and it is impossible to list them all. The following papers give basic or critical data:

Gausdal, G.; Wergeland, A.; Skavland, J.; Nguyen, E.; Pendino, F.; Rouhee, N.; McCormack, E.; Herfindal, L.; Kleppe, R.; Havemann, U.; Schwede, F.; Bruserud, Ø; Gjertsen, B.T.; Lanotte, M.; Ségal-Bendirdjian; Døskeland, S.O., *Cell Death Dis.*, **4**, e516, (2013): "Cyclic AMP Can Promote APL Progression and Protect Myeloid Leukemia Cells Against Anthracycline-Induced Apoptosis"

Jäger, R.; Russwurm, C.; Schwede, F.; Genieser, H.-G.; Koesling, D.; Russwurm, M., *J. Biol. Chem.*, **287**, 1210 - 1219 (2012): "Activation of PDE10 and PDE11 Phosphodiesterases"

Rah, S.-Y.; Mushtaq, M.; Nam, T.-S.; Kim, S.H.; Kim, U.-H., *J. Biol. Chem.*, **285**, 21877 - 21887 (2010): "Generation of Cyclic ADP-ribose and Nicotinic Acid Adenine Dinucleotide Phosphate by CD38 for Ca²⁺ Signaling in Interleukin-8-treated Lymphokine-activated Killer Cells"

Gerlo, S.; Verdood, P.; Kooijman, R., *J. Interferon Cytokine Res.*, **30**, 883 - 891 (2010): "Modulation of Cytokine Production by Cyclic Adenosine Monophosphate Analogs in Human Leukocytes"

Miki, H.; Zhou, Z.; Li, M.; Hwang, T.-C.; Bompadre, S.G., *J. Biol. Chem.*, **285**, 19967 - 19975 (2010): "Potentiation of Disease-associated CFTR Mutants by Hydrolyzable ATP Analogs"

Voisin, P.; Bernard, M., *J. Neurochem.*, **110**, 318 - 327 (2009): "Cyclic AMP-dependent Activation of Rhodopsin Gene Transcription in Cultured Retinal Precursor Cells of Chicken Embryo"

Waidmann, O.; Pleli, T.; Dvorak, K.; Baehr, C.; Mondorf, U.; Plotz, G.; Biondi, R.M.; Zeuzem, S.; Piiper, A., *J. Biol. Chem.*, **284**, 32256 - 32263 (2009): "Inhibition of the Equilibrative Nucleoside Transporter 1 and Activation of A2A Adenosine Receptors by 8-(4-Chlorophenylthio)-modified cAMP Analogs and their Hydrolytic Products"

Moll, D.; Prinz, A.; Gesellchen, F.; Drewińska, S.; Zimmermann, B.; Herberg, F.W., *J. Neural. Transm.*, **113**, 1015 - 1032 (2006): "Biomolecular Interaction Analysis in Functional Proteomics"

Meves, H., *Curr. Neuropharmacol.*, **4**, 41 - 57 (2006): "The Action of Prostaglandins in Ion Channels"

Jensen, B.O.; Selheim, F.; Døskeland, S.O.; Gear, A.R.L.; Holmsen, H., *Blood*, **104**, 2775-2782 (2004): "Protein Kinase A Mediates Inhibition of the Thrombin-Induced Platelet Shape Change by Nitric Oxide"

Kopperud, R.; Krakstad, C.; Selheim, F.; Doeskeland, S. O., *FEBS Lett.*, **546**, 121 - 126 (2003): "cAMP Effector Mechanisms. Novel Twists for an 'Old' Signaling System"

Christensen, A.E.; Selheim, F.; de Rooij, J.; Dremier, S.; Schwede, F.; Dao, K.K.; Martinez, A.; Maenhaut, C.; Bos, J.L.; Genieser, H.-G.; Døskeland, S.O., *J. Biol. Chem.*, **278**, 35394 - 35402 (2003): "cAMP Analog Mapping of Epac1 and cAMP-Kinase. Discriminating Analogs Demonstrate that Epac and cAMP-Kinase Act Synergistically to Promote PC-12 Cell Neurite Extension"

Holm, A.M.; Aukrust, P.; Aandahl, E.M.; Müller, F.; Tasken, K.; Frøland, S.S., *J. Immunol.*, **170**, 5772 - 5777 (2003): "Impaired Secretion of IL-10 by T Cells from Patients with Common Variable Immunodeficiency-Involvement of Protein Kinase A Type I"

Zhu, S.; Han, C.G.; White, R.E., *J. Vasc. Res.*, **39**, 477 - 488 (2002): "PGE₂ Action in Human Coronary Artery Smooth Muscle: Role of Potassium Channels and Signaling Cross-Talk"

Sellak, H.; Yang, X.; Cao, X.; Cornwell, T.; Soff, G.A.; Lincoln, T., *Circ. Res.*, **90**, 405 - 412 (2002): "Sp1 Transcription Factor as a Molecular Target for Nitric Oxide- and Cyclic Nucleotide-mediated Suppression of cGMP-dependent Protein Kinase-1 α Expression in Vascular Smooth Muscle Cells"

Guillemin, M.-C.; Raffoux, E.; Vitoux, D.; Kogan, S.; Soilihi, H.; Lallemand-Breitenbach, V.; Zhu, J.; Janin, A.; Daniel, M.-T.; Gourmel, B.; Degos, L.; Dombret, H.; Lanotte, M.; de Thé, H., *J. Exp. Med.*, **196**, 1373 - 1380 (2002): "In Vivo Activation of cAMP Signaling Induces Growth Arrest and Differentiation in Acute Promyelocytic Leukemia"

Wang, L.; Liu, F.; Adamo, M.L., *J. Biol. Chem.*, **276**, 37242 - 37249 (2001): "Cyclic AMP Inhibits Extracellular Signal-regulated Kinase and Phosphatidylinositol 3-Kinase/Akt Pathways by Inhibiting Rap1"

Chen, P.; Hwang, T.-C.; Gillis, K.D., *J. Gen. Physiol.*, **118**, 135 - 144 (2001): "The Relationship between cAMP, Ca²⁺, and Transport of CFTR to the Plasma Membrane"

Brophy, C.M.; Woodrum, D.A.; Pollock, J.; Dickinson, M.; Komalavilas, P.; Cornwell, T.L.; Lincoln, T.M., *J. Vasc. Res.*, **39**, 95 - 103 (2001): "cGMP-dependent Protein Kinase Expression Restores Contractile Function in Cultured Vascular Smooth Muscle Cells"

Geiger, J.; Nolte, C.; Butt, E.; Sage, S.O.; Walter, U., *Proc. Natl. Acad. Sci. USA*, **89**, 1031 - 1035 (1992): "Role of cGMP and cGMP-dependent Protein Kinase in Nitrovasodilator Inhibition of Agonist-evoked Calcium Elevation in Human Platelets"

Peters, D.J.M.; Bominaar, A.A.; Snaar-Jagalska, B.E.; Brandt, R.; Van Haastert, P.J.M.; Ceccarelli, A.; Williams, J.G.; Schaap, P., *Proc. Natl. Acad. Sci. USA*, **88**, 9219 - 9223 (1991): "Selective Induction of Gene Expression and Second-Messenger Accumulation in Dictyostelium Discoideum by the Partial Chemotactic Antagonist 8-p-chlorophenylthioadenosine 3',5'-cyclic Monophosphate"

Dostmann, W.R.G.; Taylor, S.S.; Genieser, H.-G.; Jastorff, B.; Døskeland, S.O.; Øgreid, D., *J. Biol. Chem.*, **265**, 10484 - 10491 (1990): "Probing the Cyclic Nucleotide Binding Sites of cAMP-Dependent Protein Kinase I and II With Analogs of Adenosine 3',5'-Cyclic Phosphorothioates"

Corbin, J.D.; Rannels, S.R.; Flockhart, D.A.; Robinson-Steiner, A.M.; Tigani, M.C.; Døskeland, S.O.; Suva, R.H.; Miller, J.P., *Eur. J. Biochem.*, **125**, 259 - 266 (1982): "Effect of Cyclic Nucleotide Analogs on Intrachain Site 1 of Protein Kinase Isozymes"

References cited in this Technical Information:

Singh, A.K.; Tasken, K.; Walker, W.; Frizzell, R.A.; Watkins, S.C.; Bridges, R.J.; Bradbury, N.A., *Am. J. Physiol.*, **275**, C562 - C570 (1998): "Characterization of PKA Isoforms and Kinase-dependent Activation of Chloride Secretion in T84 Cells"

Sandnes, D.; Jacobson, F.W.; M. Refsnes; T. Christoffersen, *Pharmacol. Toxicol.*, **79**, 15 - 22 (1996): "8-Bromo-cAMP and 8-CPT-cAMP Increase the Density of β -Adrenoceptors in Hepatocytes by a Mechanism Not Mimicking the Effect of cAMP"

Sugita, S.; Baxter, D.A.; Byrne, J.H., *J. Neurophysiol.*, **72**, 1250 - 1259 (1994): "cAMP-independent Effects of 8-(4-parachlorophenylthio)-cyclic AMP on Spike Duration and Membrane Currents in Pleural Sensory Neurons of Aplysia"

Connolly, B.J.; Willits, P. Barnaby; Warrington, B.H.; Murray, K.J.; *Biochem. Pharmacol.*, **44**, 2303 - 2306 (1992): "8-(4-Chlorophenyl)thio-cyclic AMP is a potent inhibitor of the cyclic GMP-specific phosphodiesterase (PDE V)"

Sandberg, M.; Butt, E.; Nolte, C.; Fischer, L.; Halbrügge, M.; Jahnsen, T.; Genieser, H.-G.; Jastorff, B.; Walter, U. *Biochem. J.*, **279**, 521 - 527 (1991): "Characterization of Sp-5,6-dichloro-1- β -D-ribofuranosylbenzimidazole-3',5'-monophosphorothioate (Sp-5,6-DCI-cBIMPS) as a Potent and Specific Activator of cyclic-AMP-dependent Protein Kinase in Cell Extracts and Intact Cells"

Øgreid, D.; Ekanger, R.; Suva, R.H.; Miller, J.P.; Døskeland, S.O., *Eur. J. Biochem.*, **181**, 19 - 31 (1989): "Comparison of the Two Classes of Binding Sites (A and B) of Type I and Type II Cyclic-AMP-dependent Protein Kinases Using Cyclic Nucleotide Analogs"

Miller, J.P.; Boswell, K.H.; Muneyama, K.; Simon, L.N.; Robins, R.K.; Shuman, D.A., *Biochemistry* **12**, 5310 - 5319 (1973): "Synthesis and Biochemical Studies of Various 8-Substituted Derivatives of Guanosine 3',5'- Cyclic Phosphate, Inosine 3',5'-"

Cyclic Phosphate, and Xanthosine 3',5'- Cyclic Phosphate"

Braumann, T.; Jastorff, B.; *J. Chromatogr.*, **350**, 105 - 118 (1985): "Physicochemical Characterization of Cyclic Nucleotides by Reversed Phase High- Performance Liquid Chromatography II. Quantitative Determination of Hydrophobicity".

Coulson, R.; Baraniak, J.; Stec, W.J.; Jastorff, B., *Life Sciences* **32**, 1489 - 1498 (1983): "Transport and Metabolism of N6- and C8-Substituted Analogs of Adenosine 3',5'-Cyclic Monophosphate and Adenosine 3',5'- Cyclicphosphorothioate by the Isolated Perfused Rat Kidney"