

# **Technical Information about 2'-AHC-cUMP-Agarose**

Update: August 30, 2023 ss



## Abbreviation:

# 2'-AHC-cUMP-Agarose

#### BIOLOG Cat. No.: A 164

**Description:** In 2'-AHC-cUMP-Agarose 2'- O- (6- Aminohexylcarbamoyl)uridine- 3', 5'- cyclic monophosphate (2'-AHC-cUMP, Cat. No. A 162) has been immobilized as an affinity ligand.

**Properties:** The second messenger cyclic UMP (cUMP, Cat. No. U 001) has been immobilized on agarose by an aminohexylcarbamoyl spacer attached to the ribose 2'-position of the ligand. The gel can be used for affinity chromatography of cUMP-responsive proteins, such as protein kinases, phosphodiesterases and others.

**Specification:** Suspension in 30 mM Na<sub>2</sub>HPO<sub>4</sub> buffer (pH 7). Ligand density: approximately 6  $\mu$ mol/ml of settled gel. UV:  $\lambda_{max}$  262 nm/suspension in glycol.

**Stability and Storage:** 2'-AHC-cUMP-Agarose has sufficient stability for chromatography at ambient temperature and does not need special care during handling or shipment. Nevertheless, for longer storage periods the gel should be kept in the refrigerator at +4 - +8°C. **Storage buffer should contain 0.1% sodium azide for prevention of microbial growth.** 

**Chromatography:** After equilibration with about 10 column volumes of starting buffer the affinity column is loaded with the protein solution, e.g. at 50-200  $\mu$ l/min. In order to elute other nucleotide-dependent proteins unspecifically bound, the column is washed, e.g. with 1 mM 5'-UMP. For elution of E. coli nucleoside diphosphate kinase it is recommended to wash the column with 10 mM ADP/20 mM MgCl<sub>2</sub>. Elution of target proteins is performed by a cyclic nucleotide gradient with cUMP up to 40 mM.

Suitable buffer systems for your special application have to be tested, but phosphate should be not optimal since one essential affinity interaction of cyclic nucleotides towards their target receptors is the cyclic phosphate.

Regeneration can be achieved by a combination of up to 100 mM cUMP and buffer salts or 8 M urea.

**Toxicity and Safety:** 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 work with it.

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

Not for drug, household or related uses!

#### Selected Reference for 2'-AHC-cUMP-Agarose:

Schwede, F.; Rentsch, A.; Genieser, H.-G., *Handb. Exp. Pharmacol.*, **238**, 307 - 337 (2017): "Medicinal Chemistry of the Noncanonical Cyclic Nucleotides cCMP and cUMP"

#### Selected Reference for 2'-AHC-cCMP-Agarose (Cat. No. A 158):

Hammerschmidt, A.; Chatterji, B.; Zeiser, J.; Schröder, A.; Genieser, H.-G.; Pich, A.; Kaever, V.; Schwede, F.; Wolter, S.; Seifert, R., *PLoS ONE*, **7**: e39848 (2012): "Binding of Regulatory Subunits of Cyclic AMP-Dependent Protein Kinase to Cyclic CMP Agarose"

# Selected References for cUMP (Cat. No. U 001):

Tal, N.; Morehouse, B.R.; Millman, A.; Stokar-Avihail, A.; Avraham, C.; Fedorenko, T.; Yirmiya, E.; Herbst, E.; Brandis, A.; Mehlman, T.; Oppenheimer-Shaanan, Y.; Keszei, A.F.A.; Shao, S.; Amitai, G.; Kranzusch, P.J.; Sorek, R., *Cell*, **184**, 5728 - 5739 (2021): "Cyclic CMP and Cyclic UMP Mediate Bacterial Immunity Against Phages"



# - LIFE SCIENCE INSTITUTE -

Bähre, H.; Hartwig, C.; Munder, A.; Wolter, S.; Stelzer, T.; Schirmer, B.; Beckert, U.; Frank, D.W.; Tümmler, B.; Kaever, V.; Seifert, R., *Biochem. Biophys. Res. Commun.*, **460**, 909 - 914 (2015): "cCMP and cUMP Occur in Vivo"

Beste, K.Y.; Spangler, C.M.; Burhenne, H.: Koch, K.-W.; Shen, Y.; Tang, W.; Kaever, V.; Seifert, R., *PLoS ONE*, **8**: e70223 (2013): "Nucleotidyl Cyclase Activity of Particulate Guanylyl Cyclase A: Comparison with Particulate Guanylyl Cyclases E and F, Soluble Guanylyl Cyclase and Bacterial Adenylyl Cyclases Cyaa and Edema Factor"

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

Zong, X.; Krause, S.; Chen, C.-C.; Krüger, J.; Gruner, C.; Cao-Ehlker, X.; Fenske, S.; Wahl-Schott, C.; Biel, M., *J. Biol. Chem.*, **287**, 26506 - 26512 (2012): "Regulation of Hyperpolarization-Activated Cyclic Nucleotide-Gated (HCN) Channel Activity by cCMP"

Göttle, M.; Dove, S.; Kees, F.; Schlossmann, J.; Geduhn, J.; König, B.; Shen, Y.; Tang, W.-J.; Kaever, V.; Seifert, R., *Biochemistry*, **49**, 5494 - 5503 (2010): "Cytidylyl and Uridylyl Cyclase Activity of Bacillus anthracis Edema Factor and Bordetella pertussis CyaA"

Scott, S.-P.; Shea, P.W.; Dryer, S., *Biochemistry*, **46**, 9417 - 9431 (2007): "Mapping Ligand Interactions with the Hyperpolarization Activated Cyclic Nucleotide Modulated (HCN) Ion Channel Binding Domain Using Soluble Construct"

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

Blecher, M.; Ro'Ane, J.T.; Flynn, P.D., *Arch. Biochem. Biophys.*, **142**, 351 - 362 (1971): "Biological Roles for 3',5'-Cyclic Nucleotides. I. Lipolytic Agents in Isolated Rat Epididymal Adipose Cells and Substrates for Adipose Tissue Phosphodiesterase"