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Pastan I & Perlman R. Cyclic adenosine monophosphate in bacteria. *Science* 169:339-44, 1970.
[Molec. Biol. Sect., Endocrinol. Branch, Natl. Cancer Inst., and Diabetes Sect., Clin. Endocrinol. Branch, Natl. Inst. Arthritis and Metabolic Dis., NIH, Bethesda, MD]

This paper reviewed the experiments that showed how cyclic AMP controls transcription in *Escherichia coli* and that elucidated the mechanism by which glucose represses the synthesis of a variety of inducible enzymes. [The SCI® indicates that this paper has been cited in over 420 publications since 1970.]

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"In 1961, I arrived at the National Institutes of Health (NIH) and began to work in Ed Rall's department with Jim Field on the effects of thyroid-stimulating hormone (TSH) on the thyroid gland. Later, after a postdoctoral stay in Earl Stadtman's laboratory, I returned to my studies on TSH action. The discovery by Earl Sutherland[1] that many hormones activated adenylate cyclase and thus increased intracellular cyclic AMP levels prompted me to investigate cyclic AMP in the thyroid. I found that TSH activated adenylate cyclase in thyroid and that the addition of a cyclic AMP analog to the thyroid tissue reproduced many of the actions of TSH, suggesting cyclic AMP as the mediator of TSH action. A more fundamental problem was how cyclic AMP acted, and it occurred to me that a solution might come more readily from investigations on *E. coli*. Robert Perlman and I then joined forces to study this. Sutherland had shown that the addition of glucose to glucose-starved *E. coli* lowered cyclic AMP levels in the cells. One well-known effect of glucose in *E. coli* was to repress the synthesis of a variety of inducible enzymes. Therefore, Perlman and I began to examine the hypothesis that the role of cyclic AMP in *E. coli* was to stimulate the expression of a set of genes, and that glucose acted by regulating cyclic AMP levels. We were soon able to demonstrate that cyclic AMP not only stimulated the synthesis of some enzymes known to be under glucose control but also overcame glucose repression of these enzymes.

"We submitted a paper to a rapid-publication journal but it was rejected, as were papers to two other well-known journals. Finally, a paper submitted to *Biochemical and Biophysical Research Communications*[2] was accepted, as was a paper to the *Journal of Biological Chemistry*.[3] We breathed a sigh of relief, because by this time our unpublished results were well known, the early experiments were not hard to do, and many scientists were studying the same problem. Initially, it seemed possible that cyclic AMP might regulate gene expression in a very indirect manner, but a number of subsequent observations suggested that its action might be direct. These observations included our isolation of mutants that either could not produce cyclic AMP because of a defect in adenylate cyclase (*cya*) or could not respond to cyclic AMP because of a defective cyclic AMP receptor protein (CRP); our demonstration that *lac* promotor mutants were unresponsive to cyclic AMP;[4] and the demonstration by Zubay *et al.*[5] that cyclic AMP had an action in the cell-free synthesis of β-galactosidase. Finally, we were able to purify CRP and demonstrate in a test-tube reaction containing defined components (cyclic AMP, CRP, RNA polymerase, *lac* DNA) that transcription of the *lac* operon required cyclic AMP and CRP and was inhibited by purified *lac* repressor.[6,7] This was the first experiment that demonstrated the expression of a bacterial gene and its regulation using pure components.

"These studies provided direct confirmation of the regulatory models proposed by Jacob and Monod[8] and serve today as a model of gene regulation. I believe that our initial discoveries would have been more readily accepted and published if they had not contradicted the orthodox belief of how catabolites of glucose controlled gene expression. My fruitful collaboration with Perlman (now at the University of Illinois) and our joint excitement about the implications of our efforts in the newly emerging field of molecular biology kept us working with enthusiasm even though we had difficulty publishing our initial studies."

1. **Sutherland E W.** Studies on the mechanism of hormone action. *Les Prix Nobel.* Stockholm: Norstedt & Söner, 1972. p. 240-57.
2. **Perlman R & Pastan I.** Cyclic 3',5'-AMP: stimulation of galactosidase and tryptophanase induction in *E. coli*. *Biochem. Biophys. Res. Commun.* 30:656-64, 1968. (Cited 165 times.)
3. ----------. Regulation of β-galactosidase synthesis in *Escherichia coli* by cyclic adenosine 3',5' monophosphate. *J. Biol. Chem.* 243:5420-7, 1968. (Cited 175 times.)
4. **Pastan I & Perlman R.** The role of the *lac* promotor locus in the regulation of β-galactosidase synthesis by cyclic 3',5'-AMP. *Proc. Nat. Acad. Sci. US* 61:1336-42, 1968. (Cited 70 times.)
5. **Chambers D A & Zubay G.** The stimulatory effect of cyclic adenosine 3'5'-monophosphate on DNA-directed synthesis of β-galactosidase in a cell-free system. *Proc. Nat. Acad. Sci. US* 63:118-22, 1969. (Cited 105 times.)
6. **Anderson W B, Schneider A B, Emmer M, Perlman R L & Pastan I.** Purification of and properties of the cyclic adenosine 3',5'-monophosphate receptor protein which mediates cyclic AMP dependent gene transcription in *Escherichia coli*. *J. Biol. Chem.* 246:5929-37, 1971. (Cited 150 times.)
7. **de Crombrugghe B, Chen B, Anderson W, Nissley P, Gottesman M, Pastan I & Perlman R.** *Lac* DNA, RNA polymerase and cyclic AMP receptor protein, cyclic AMP, *lac* repressor and inducer are the essential elements for controlled *lac* transcription. *Nature—New Biol.* 231:139-42, 1971. (Cited 140 times.)
8. **Jacob F & Monod J.** Genetic regulatory mechanisms in synthesis of proteins. *J. Mol. Biol.* 3:318-56, 1961. (Cited 2,740 times.)