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Inhibition of conidiation and photo-induced carotenoid biosynthesis by cyclic-AMP

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Abstract

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Cyclic-AMP (adenosine 3':5'-cyclic-monophosphoric acid) has been found in a wide variety of organisms. Regulation of enzyme systems by this compound has been shown in some cases to be due to an effect on the transcription of genes, on the translation of m-RNA, or on the activity of enzymes (Jost and Rickenberg 1971 Ann. Rev. Biochem. 40: 741). In Neurospora cyclic-AMP has been shown to derepress tyrosinase and L-amino acid oxidase (Feldman 1969 Biology Ann. Rept., Calif. Inst. Technology, p. 160). Also, the rate

of conversion of the inactive form of glycogen phosphorylase to the active form has been shown to be increased by cyclic-AMP in this organism (Tellez-Iñon and Torres 1970 Proc. Nat. Acad. Sci. U. S. A. 66: 459). Adenylcyclase, the enzyme which catalyzes the synthesis of cyclic-AMP, has been isolated from Neurospora (Flawia and Torres 1972 J. Biol. Chem. 247: 6873).

It was decided to determine whether exogenously added cyclic-AMP has any effect on the photoinduction of carotenoid synthesis in N. crassa. Cultures were grown for 6 days at 18°C in 20 ml portions of Vogel's minimal medium supplemented with 0.8% Tween-80. Under red safe-light (General Electric-BCJ), the excess medium was removed from the mycelial pads with suction on a Buchner funnel, and they were then placed in petri dishes, 3 pads per dish. 2 ml aliquots of the cyclic-AMP solutions to be tested (cyclic-AMP in Vogel's minimal medium supplemented with 0.8% Tween-80) were added to each pad. The pads were incubated in the dark for 2 hours at 25°C and then irradiated continuously for 6 hours at 25°C using Westinghouse cool-white fluorescent lamps at an intensity of 200 foot-candles. The carotenoid pigments were extracted from the pads and assayed using standard technique.

Carotenoid synthesis was found to be inhibited by cyclic-AMP. The percent inhibition measured was 60% for 20mM cyclic AMP, 15% for 10mM and 0% for 5 mMolar. Photoinduction of carotenoid synthesis in N. crassa can be divided into at least three distinct sequential phases: (1) a rapid light reaction, (2) a period of protein synthesis, and (3) accumulation of the carotenoid pigments (Harding and Mitchell 1968 Arch. Biochem. Biophys. 128: 814). It is planned to investigate whether cyclic-AMP inhibits the accumulation of pigment by blocking the light reaction, protein synthesis or some unknown step in the photoinduction process. If the irradiation were continued for a total time of 24 hours, it was observed that conidiation was completely inhibited by 20 and 10 mM solutions of cyclic-AMP and partially inhibited by 5 mM solutions. Elucidation of the basis for this effect may eventually yield information about the mechanism of conidiation. It should be noted that addition of cyclic-AMP to Vogel's minimal medium causes a pH decrease. For a 20 mM solution, the pH is decreased from 5.8 to 4.2. Appropriate controls were carried out to show that the inhibitory effects described above were not due to a pH change.

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