the hypolipidemic property of these drugs and not to independent actions. In addition, to our knowledge, all known peroxisome proliferators have hypolipidemic properties including acetylsalicylic acid, which produces a minimal to moderate increase in peroxisome profiles (*22*). The mechanism by which these hypolipidemic drugs produce peroxisome proliferation in the liver cells and their role in lipid metabolism are not understood. The frequent association of hepatic peroxisome proliferation with drug-induced hypolipidemia suggests that either peroxisome catalase or some other peroxisomal enzyme may be responsible for the hypocholesterolemic and hypotriglyceridemic effects (*11, 23*). However, only if a nonhypolipidemic peroxisome proliferator is found can these two effects be considered unrelated. Until such a compound is identified, it is reasonable to direct future studies towards clarification of the role of peroxisomes in lipid metabolism.

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1 April 1975

Circadian Periodicity in *Neurospora*: Alteration by Inhibitors of Cyclic AMP Phosphodiesterase

Abstract. *Three methyl xanthine inhibitors of adenosine 3',5'-monophosphate phosphodiesterase—theophylline, aminophylline, and caffeine—lengthen the period of the circadian conidiation rhythm of* Neurospora. *The effects are seen in wild-type strains and in three mutant strains with genetically altered period lengths. These results suggest the possible involvement of adenosine 3',5'-monophosphate in the control of circadian rhythmicity.*

Recent interest in the molecular basis of circadian rhythmicity has focused on the possible role of membranes in this phenomenon. Much of the impetus for this thinking has come from experiments in which substances likely to affect membrane structure or function alter circadian periodicity. For example, ethyl alcohol and D_2O cause phase shifts or changes of period length in the leaf movement rhythm of *Phaseolus* (*1*), in the activity rhythm of the isopod *Excirolana* (*2*), and the phototaxis rhythm of *Euglena* (*3*). In addition, pulses of K^+ cause phase shifts in the spontaneous rhythmic firing of the optic nerve of *Aplysia* (*4*), and valinomycin, which alters membrane transport of K^+, causes phase shifts in *Phaseolus* (*5*) and *Gonyaulax* (*6*). Much of this type of information has been incorporated in the ion-flux model of the circadian clock proposed by Njus *et al.* (*7*).

Adenosine 3',5'-monophosphate (cyclic AMP) is ubiquitous in both higher and lower organisms and plays a variety of regulatory roles in these systems (*8*). Adenylate cyclase and cyclic AMP phosphodiesterase, the enzymes that control endogenous cyclic AMP levels, are often mem-

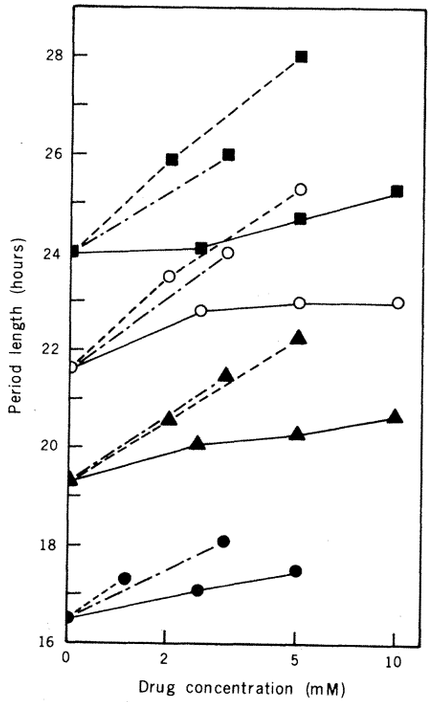

Fig. 1. Lengthening the period of the circadian rhythm of conidiation in *Neurospora crassa*. Each line represents one strain tested with one drug at the indicated concentrations. Symbols used to denote the various strains are as follows: ○, *frq*$^+$; ●, *frq-1*; ▲, *frq-2*; and ■, *frq-3*. Symbols used to denote different drugs are as follows: ———, theophylline; ------, aminophylline; and — — —, caffeine. Each point represents the mean of 12 replicate growth tubes. In all but three cases, the standard deviations ranged from 0.2 to 0.6. In the other three cases they ranged from 0.7 to 0.9.

brane-bound, and their activities are affected by many of the same substances that affect circadian rhythms (*9*). In addition, recent measurements on mammalian cells in culture have demonstrated fluctuations in cyclic AMP levels associated with the cell cycle (*10*) and have suggested the possibility of an oscillatory feedback system in which the levels of cyclic AMP itself affect the synthesis of these two enzymes or their activities (*11*). In view of the possible relation between circadian clock cycles and cell cycles (*12*), oscillations in cyclic AMP levels offer an attractive possibility for a role in the control of circadian clocks. This report presents our initial attempts to determine whether cyclic AMP is involved in the circadian clock of *Neurospora* and shows a lengthening of the period of the circadian conidiation rhythm by three methyl xanthine inhibitors of cyclic AMP phosphodiesterase.

The following strains of *Neurospora crassa* were used in these experiments: *band*, which serves as the "wild type" (*13*), and three mutants derived from *band*, each of which has a period length different from *band*: *frq-1*, *frq-2*, and *frq-3* (*14*). All culture conditions, media, and procedures for measuring period length of the conidiation rhythm on "race" tubes were as previously described (*14*).

Theophylline, an inhibitor of *Neurospora* cyclic AMP phosphodiesterase (*15*), causes a small increase in the period lengths of the conidiation rhythm in wild-type and in each of the three mutant strains (see Fig. 1). Aminophylline, a derivative of theophylline that enters the cells more readily than theophylline and therefore inhibits phosphodiesterase in vivo at lower concentrations than theophylline (*16*), also causes a significant period lengthening and, as expected, at lower concentrations than theophylline. Finally, caffeine, another inhibitor of cyclic AMP phosphodiesterase (*15*), also causes significant increases in period length (Fig. 1).

Although at high concentrations all of these compounds are toxic to the organism, period lengthening occurred at concentrations that correspond to those necessary to inhibit phosphodiesterase (*15*) and that did not inhibit growth rate. In fact some low concentrations actually caused a small increase in linear growth rate (Table 1), a phenomenon similar to that previously reported in *Neurospora* (*16*).

These results indicate that inhibitors of cyclic AMP phosphodiesterase lengthen the period of the *Neurospora* clock. In addition, at least one previous report (*17*) demonstrated a period lengthening effect of theophylline and caffeine in higher plants, although their relation to cyclic AMP metabolism was not recognized at that time.

Table 1. Effect of aminophylline on linear growth rate of *Neurospora*. All standard deviations were 0.2 cm or less.

Strain	Linear growth rates (cm/day) at the following concentrations (m*M*) of aminophylline			
	0	1	2	5
frq$^+$	2.51	2.69	3.07	2.74
frq-1	2.48	2.49	3.04	2.70
frq-2	2.57	2.66	3.08	2.72
frq-3	2.46	2.71	2.91	2.74

It would be premature to conclude, however, that such results demonstrate that cyclic AMP is involved directly in the control of the circadian clock. First of all, it is not known definitely that the effect of the drugs is mediated through their inhibition of phosphodiesterase, since they are known to affect other cellular processes, such as ion transport and macromolecular synthesis. However, the effectiveness of all three inhibitors at concentrations known to inhibit *Neurospora* phosphodiesterase and at concentrations that do not inhibit growth tend to reduce, although do not eliminate, this possibility. Even if the inhibitors are acting through phosphodiesterase to change the cyclic AMP levels, it cannot yet be inferred that cyclic AMP metabolism is part of the clock, for this can be done only when changes in endogenous cyclic AMP levels can be correlated quantitatively with changes in the overt behavior of the clock.

Nevertheless, it is interesting that one important property of circadian clocks—namely, sensitivity to light—could be accounted for in terms of cyclic AMP metabolism. In both isolated frog retinal tissue (*18*) and the fungus *Phycomyces* (*19*) visible light causes a significant decrease in the endogenous cyclic AMP levels, and at least for frog retinal tissue, this effect is due to the light activation of phosphodiesterase. This type of phenomenon is exactly what has been predicted in several molecular models for the clock (*20*).

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21. Supported by NIH grants GM-19850 and GM-22144. I thank Marian Hoyle for her excellent assistance.

9 May 1975; revised 30 July 1975

Immunosurveillance of Naturally Occurring Feline Leukemia

Abstract. *When compared to their housemates that subsequently developed leukemia, cats that remained healthy had five- to tenfold higher (geometric mean) humoral antibody titers to the feline oncornavirus-associated cell membrane antigen. This is compatible with the application of the immunosurveillance hypothesis to the natural development of leukemia in an outbred mammalian species.*

People with either the genetic immunologic deficiency diseases or drug-induced immunosuppression have an increased risk for developing lymphoid leukemia or lymphoma (*1*). This type of observation led Burnet to propose that an immunosurveillance mechanism may operate under natural conditions to eliminate malignant cells (*2*). Most studies with inbred mice and other laboratory rodents support this hypothesis (*3*). Some observations, however, such as the apparent lack of an increased risk for development of some types of tumors in genetically athymic (nude) mice have provided arguments against the immunosurveillance concept (*4*). It has been difficult to design experiments that test the application of the immunosurveillance hypothesis to spontaneous neoplasms of outbred mammals because the etiologic agents for naturally occurring tumors are generally unknown,