shown in figure 2. The diaphragmatic glycogen levels of lard-fed rats were much lower than those of control, and the 24-h fluctuation was much less dramatic. Also illustrated in figure 2 is the circadian rhythm of hepatic glycogen in chow-fed rats. Hepatic glycogen content in lard-fed rats diminished over most of the 24-h cycle, and was not statistically rhythmic. At 16.00 and 20.00 h, hepatic glycogen levels of chow-and lard-fed rats were not different.

The entrainment of feeding activity of both animal groups to the light: dark cycle is apparent from the stomach contents data shown in figure 1. Although the feeding patterns of chow-fed and lard-fed rats are identical, there are differences in the times of the glycogen peaks and nadirs between the 2 diet groups and among the 3 tissues. The volume of food ingested by the lard-fed rats was lower than controls at all times sampled due to the low bulk and high caloric content of the high lipid diet.

This study confirms a differential effect of a lipid diet on ventricular and diaphragmatic glycogen rhythms and indicates a difference in metabolic control mechanisms between these 2 types of continuously active muscles. The increase in ventricular glycogen in lipid-fed rats presumably results from an acceleration of fatty acid oxidation which causes an inhibition of glycolysis at the phosphofructokinase[15] and pyruvate dehydrogenase[16] enzymes. The production of $^{14}CO_2$ from labeled pyruvate and pyruvate uptake have been shown to decline in diaphragms of rats fed high fat diets, even though acetate oxidation was unchanged, implying a blockage of glycolysis at pyruvate dehydrogenase[10,11]. There seems to be no evidence of a similar blockage at phosphofructokinase in diaphragm with lipid feeding. Diaphragm, unlike heart, does not have significant glycerol kinase activity[17], and utilizes the glycolytic production of dihydroxyacetone-phosphate to provide glycerol phosphate for the synthesis of triglyceride. The inhibition of glycolysis at pyruvate dehydrogenase, but not at phosphofructokinase, would allow the formation of dihydroxyacetonephosphate as a source of glycerol-phosphate, and help to explain a decline in diaphragm glycogen in response to a diet that is known to increase diaphragm triglycerides[11].

The depletion of liver glycogen in the lard-fed rats is consistent with the role of the liver in the maintenance of blood glucose during carbohydrate deprivation. However, it has been reported that rat liver phosphorylase is inhibited by feeding a fat diet[18], and the decline in glycogen levels may be due as well to a decrease in glycogen synthesis. Without cyclic fluctuations in the carbohydrate delivered to the liver from absorbed nutrients, the rhythmicity of liver glycogen is lost or significantly dampered. It is apparent from this study that the 24-h variation in liver glycogen in control rats is quite dramatic.

The comparison of ventricular and hepatic glycogen in chow-fed and lard-fed rats would have yielded no differences in tissue levels if sampling were limited to certain times in the circadian cycle. This point is germane to any study of metabolism in tissues which have substrate rhythms.

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Behavioral fever induced in guinea-pigs by intrapreoptic pyrogen[1]

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Summary. The preoptic area of the hypothalamus integrates not only the autonomic, but also the behavioral components of fever in guinea-pigs.

It is now generally recognized that behavioral adjustments are an integral part of physiological temperature regulation. It is not surprising, therefore, that behavioral responses also are normal adjuncts of fever production and lysis in all classes of vertebrates, from fishes to mammals[2]. A commonplace example among mammals is man in the first stages of fever reporting feeling cold, seeking increased insulation and reducing his heat-exchanging surface area in order to warm himself. Fever also has a great influence on thermal preference. Thus, fishes and amphibians actively seek higher ambient temperatures following injections of pathogenic organisms; and, in operant selection of thermal reinforcers experiments, cats, dogs, and baboons injected systemically with pyrogens respond with an increased rate of lever-pressing for external heat during the phase of rising temperature.

It has been established that the preoptic-anterior hypothalamic (POAH) region is involved in the control of behavioral thermoregulation[3]. Thus, warming this area elicits a decrease in behavioral heat-acquisition responses, an increase in behavioral heat-escape responses, and a lowering of an animal's preferred ambient temperature; cooling this area produces the opposite effects. That the POAH also is the locus of greatest control of autonomic thermoregulatory responses has been repeatedly shown[4]. Since endogenous pyrogen (EP) also localizes in this region to trigger the total

pattern of autonomic fever-producing mechanisms[5-10], this study was untertaken to determine whether the POAH similarly activates EP-induced behavioral fever.

Thermoregulatory behavior was assessed in these experiments by monitoring the thermopreferendum of adult guinea-pigs in a thermocline of local fabrication, consisting of a triple-walled, clear plexiglass, 1-m-long cylinder. A thermal gradient of 10 °C was established over its length, from air inlet to outlet, by regulating the flow rate and the temperature of water and air flowing counter-currently, viz., cool water was continuously circulated through the inner of the 2 jackets of this chamber while warm air entered the innermost cylinder which contained the animal. Air and wall temperatures were measured by means of thermistors installed at 12-cm intervals in the chamber; the data were multiplexed to the input of a telethermometer, the output of which, in turn, was formated by a Mostek KIM-1 Microprocessor (Commodore Business Machines, Palo Alto, CA) and digitized for print-out on a teletypewriter. The animal's location within the chamber was detected by its breaking of one or more far-red light beams directed toward 8 photoelectric cells located also at 12-cm intervals in the wall of a platform upon which the guinea-pig was completely free to move. The output of the 8 photoresistors passed through a parallel-to-serial converter and was fed into the microprocessor, which, in turn, modulated it for recording at 1-min intervals on the teletypewriter. Only the signal from the light beams interrupted by an animal in their path was printed.

Body temperatures were measured using a precalibrated, temperature-sensitive, wirless, miniature radiotransmitter (Model X, Mini-Mitter Co., Indianapolis, IN) which emitted a series of pulses at a frequency between 1 and 2 Hz, the rate being proportional to the temperature of the probe's environment. The signal was received on the AM band of an ordinary radio, and thus rendered audible as clicks. To obtain a visible record of the clicks, the output from the radio was fed to a locally fabricated 'click receiver' printed-circuit board, and from there into the KIM-1 Microprocessor. The clicks were accumulated for 1 min, then fed out through a digital-to-analog converter to the teletypewriter. This transmitter was implanted s.c. into the interscapular space of each guinea-pig.

6 adult, male, English short-haired guinea-pigs, ranging in weight from 500 to 700 g, were used in these experiments. In separate runs, the guinea-pigs were placed into the thermocline (which was covered with a small dropcloth to obviate environmental distractions which might affect the animals' behavior) and allowed to move freely. For the first 90 min of this exposure, the chamber was maintained at room temperature without a thermal gradient; for the following 2 h, a gradient from 27 to 38 °C was established. No treatments were given. Under these conditions, the animals moved randomly within the thermocline when no gradient was present; their body temperature (T_{bo}) was 37.5 ± 0.4 °C (SD). They selected a mean ambient temperature (T_a) of 29.0 ± 0.4 °C after approximately 40 min when the gradient was present; their T_{bo} stabilized at 38.0 ± 0.3 °C in this environment.

Beginning 3 days later, the animals, in separate ex-

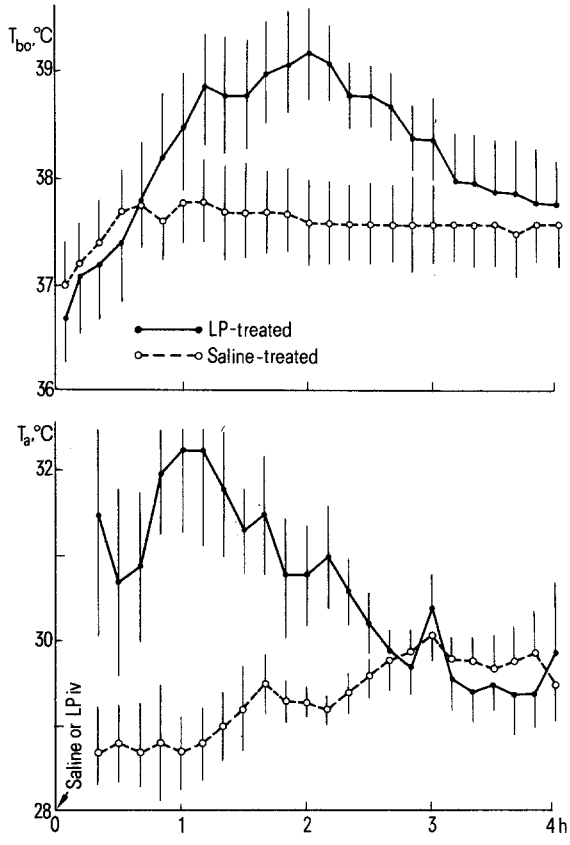

Fig. 1. Body (T_{bo}, upper panel) and preferred ambient (T_a, lower panel) temperatures of conscious, free-moving guinea-pigs following the i.v. injection at time 0 of 1.0 ml of either apyrogenic saline (dashed lines) or leukocytic pyrogen (LP, solid lines). The vertical lines indicate the SD of the means.

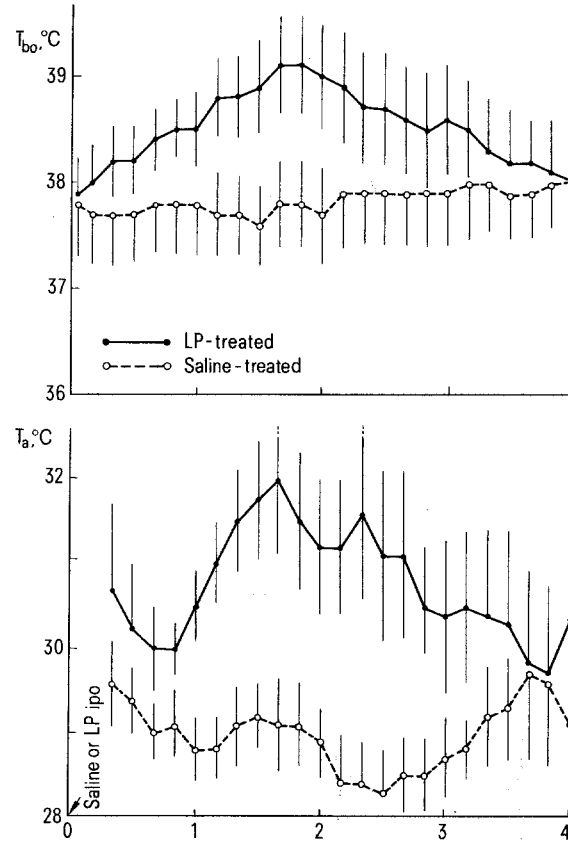

Fig. 2. Body (T_{bo}, upper panel) and preferred ambient (T_a, lower panel) temperatures of conscious, free-moving guinea-pigs following the bilateral intrapreoptic (ipo) injection of 1.0 µl of either apyrogenic saline (dashed lines) or leukocytic pyrogen (LP, solid lines). The vertical lines indicate the SD of the means.

periments, were lightly anesthetized with ether and injected into an ear vein, randomly, with 1.0 ml of either apyrogenic saline or leukocytic pyrogen (LP)[11]. Upon regaining consciousness (usually in less than 10 min), the animals were placed into the cool end of the thermocline (again end-to-end gradient from 27 to 38 °C) and allowed to seek their preferred temperature for the next 4 h. The results are illustrated in figure 1. The T_{bo}s of the saline-injected guinea-pigs stabilized at 37.8 ± 0.4 °C after 40 min. By contrast, those of the LP-treated animals rose rapidly for the first 70 min, and then very slowly during the following 50 min, essentially stabilizing. After 120 min, they gradually declined. Defervescence was completed in 240 min post-LP injection. Over the 4 h of their exposure to the thermocline, the saline-treated animals gradually moved toward warmer T_as, from 28.7 ± 0.5 to 29.7 ± 0.4 °C. By contrast, the LP-treated guinea-pigs, coincidentally with the rising of their fever, sought a significantly warmer environment than the control animals during the first 70 min. During the following 50 min, i.e., during the period of high stable T_{bo}, they selected progressively lower T_as. During defervescence, they moved more rapidly to even cooler temperatures, and by the end of the experiment, they were at T_as not different from those of the controls.

Beginning 2 days after these experiments, each of the above animals was anesthetized with sodium pentobarbital and guide cannulas were implanted under stereotaxic guidance bilaterally into the preoptic area; the position of the cannula tips was verified histologically at the conclusion of these experiments[10]. 4 days after this surgery, each animal again was placed into the cool end of the thermocline (end-to-end gradient as before from 27 to 38 °C) to run freely for 2 h; no injections were given. Under these conditions, the animals selected 29.1 ± 0.4 °C after 40 min of random activity; T_{bo} averaged 38.0 ± 0.2 °C.

On subsequent days, 1.0 µl of either apyrogenic saline or LP, in separate randomized experiments, was injected bilaterally into the preoptic area of each guinea-pig, without anesthesia. The animal was then immediately placed into the cool end of the thermocline (gradient as before), and allowed to seek its preferred temperature for the next 4 h. T_{bo}, T_a, and animal location were monitored as before. The results are shown in figure 2. While the T_{bo}s of the saline-treated guinea-pigs remained stable at 37.8 ± 0.5 °C throughout the 240-min exposure, those of the LP-treated animals increased immediately, reaching their fastigium at 100 min. Defervescence began at 110 min and was completed by 240 min. The control guinea-pigs remained in 29.0 ± 0.4 °C throughout their exposure to the thermocline; by contrast, from 50 min post-LP onward, the LP-treated animals chose increasingly warmer T_as, coincidentally with the rising phase of their fever. They selected their highest T_a at 100 min, then gradually moved again toward cooler temperatures.

Hence, the present results would permit the conclusion that the preoptic area integrates not only the autonomic, as shown earlier[10], but also the behavioral components of fever in adult guinea-pigs.

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The mechanical and biochemical effects of pentoxifylline on the perfused rat heart[1]

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Summary. Perfusion of the isolated rat heart at constant heart rate and coronary flow with the inhibitor of cyclic nucleotide phosphodiesterase, pentoxifylline (10^{-4} moles/l), produced no significant effect on the maximum rate and the peak of contraction, but increased the maximum rate of relaxation. cAMP level and cAMP-dependent protein kinase activity were increased in the absence of changes in cGMP. The results were identical in hearts of reserpinized rats.

Increases in cardiac contractility are associated with increased intracellular levels of adenosine 3′:5′ cyclic monophosphate (cAMP)[3]. Catecholamines that stimulate myocardial β-adrenergic receptors produce their positive inotropic response by increasing cAMP production[4]. Myocardial cAMP is also increased by inhibition of its breakdown to 5′ AMP by phosphodiesterase inhibition[5–7]. The increase in intracellular concentration of cAMP is thought to activate a cAMP-dependent protein kinase which in turn phosphorylates specific cellular proteins, such as proteins of myocardial sarcolemma[8], the contractile protein troponin[9] and the phospholamban in the sarcoplasmic reticulum[10]. This protein phosphorylation might lead to changes in contractility and relaxation through alteration of Ca^{++} movement. Protein kinase-dependent phosphorylation of phospholamban should result in an enhancement of Ca^{++} transport into the sarcoplasmic reticulum[11] causing accelerated relaxation[12]. An accelerated relaxation has been detected in our laboratory with isoproterenol and was mimicked by dibutyryl 3′:5′-cAMP[13].

The possibility that guanosine 3′:5′ cyclic monophosphate (cGMP) may have regulatory actions on the heart antagonistic to those of cAMP has been reported[14]. Cholinergic agents have been shown to increase myocardial cGMP level and to decrease contractile force[15].

The reported inotropic and relaxant effects of phosphodiesterase inhibitors are controversial[3,6,16–20]. The present study was undertaken to provide information about the mechanical behavior of the perfused rat heart under the effect of the phosphodiesterase inhibitor, pentoxifylline, and its relation with myocardial cAMP-dependent protein kinase activity, cAMP and cGMP levels.