

oxygen tension than their myocardial counterparts and could, therefore, exert a finer control in regulation of coronary blood flow. We have shown (6) that the 5'-nucleotidase from rat myocardium is strongly inhibited by low concentrations of ATP (K_i 1.83×10^{-6} M). It was suggested that during oxygen sufficiency, the enzyme may be kept in restraint by virtue of high intracellular ATP concentrations. During hypoxia, when ATP levels decline, adenosine formation would be facilitated by removal of inhibition. Possible similarities between the myocardial enzyme and the present activity in coronary vasculature are clearly of interest.

It has been assumed that the coronary dilator action of ATP and 5'-AMP is due to their prior conversion to adenosine (9, 10), which readily crosses cell membranes. The rapid conversion of ATP and 5'-AMP to adenosine by perfused hearts as described here would lend strong support to this idea.

Summary. ATP and 5'-AMP were extensively degraded to adenosine and inosine during a single passage through rat hearts

perfused in an open-flow system. It is suggested that enzymes which convert adenine nucleotides to adenosine exist within, or on, coronary vascular cells. Whether these enzymes contribute to the formation of adenosine during cardiac hypoxia is unknown.

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Changes in Liver Lipid Composition with Overnight Fasting* (32615)

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We previously observed (1) that fasting for 24 hours resulted in a 34% loss of liver weight, a 19% loss in liver phospholipid, and a significant decrease in the relative proportion (wt. %) of oleate in total liver fatty acids, together with small increases in linoleate and arachidonate. These results indicated a need to study the extent of changes which might result from fasts shorter than 24 hours, such as "overnight," since these intervals have been used in studies of lipid composition (2-4). The following experiment compared the fatty acid composition of liver lipid fractions from rats fasted overnight (ca.

12 hours) with the values from fed rats, as well as with a group fasted for 22 hours.

Materials and Methods. Male weanling Long-Evans rats were caged individually in suspended, galvanized screen-wire cages. They had been fed a 20% casein-6% cottonseed oil-sucrose diet (5) *ad libitum* for 6 weeks, since this is a period frequently used in nutritional studies with rats fed purified diets. The rats were divided into 4 groups: (a) fed, killed at 8:30 a.m.; (b) fasted from 8 p.m. to 8:30 a.m.; (c) fasted from 8 p.m. to 6 p.m.; (d) fed, killed at 8 p.m., at the start of the fasting period for group b. The average initial weight for all groups ranged from 288 to 296

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gm. Group b lost 12 gm and group c lost 17 gm by the end of the fasting period. Group a ate 16 gm food (range 13-18 gm) and gained 6 gm. The *ad libitum* food intake of all groups was 18-22 gm in the 24-hour period (8 p.m. to 8 p.m.) before the experiment. At the time of autopsy, the rats were anesthetized with amobarbital (Sodium Amytal) and bled by heart puncture. Livers were removed, weighed, and lyophilized. Liver lipids were extracted and analyzed by methods previously used (6,7).

Results and Discussion. The liver weights

TABLE I. Phospholipid Fatty Acid Composition.

Group	Phospholipid		Phospholipid fatty acids (wt. %)						
	Liver wt. (gm)	(mg/liver)	(mg/gm)	16:0	16:1	18:0	18:1	18:2	20:4
a (13) ^a	12.9 ± 0.7	341 ± 15	26.5 ± 0.7	19.0 ± 0.3	0.8 ± 0.1	25.3 ± 0.5	6.2 ± 0.2	13.6 ± 0.5	34.2 ± 0.6
b (13)	9.2 ± 0.2	318 ± 9	34.7 ± 0.6	18.0 ± 0.4	0.4 ± 0.1	27.8 ± 0.5	4.3 ± 0.1	10.8 ± 0.3	38.4 ± 0.4
c (6)	8.2 ± 0.3	283 ± 6	34.5 ± 0.7	17.4 ± 0.7	0.4 ± 0.1	27.9 ± 0.7	4.4 ± 0.1	11.1 ± 0.7	38.4 ± 0.4
d (14)	11.4 ± 0.4	350 ± 13	30.6 ± 0.6	18.1 ± 0.3	0.5 ± 0.1	27.3 ± 0.2	5.3 ± 0.2	11.6 ± 0.4	36.2 ± 0.2

^a Number in parentheses is the number of rats per group. Group a: fed, killed at 8:30 a.m.; Group b: fasted from 8 p.m. to 8:30 a.m.; Group c: fasted from 8 p.m. to 6 p.m.; Group d: fed, killed at 8 p.m. at start of fasting period for Group b.

TABLE II. Triglyceride Fatty Acid Composition.

Group	Triglyceride		Triglyceride fatty acids (wt. %)						
	Liver wt. (gm)	(mg/liver)	(mg/gm)	16:0	16:1	18:0	18:1	18:2	20:4
a (13) ^a	12.9 ± 0.7	101 ± 8	7.8 ± 0.5	32.8 ± 1.7	5.2 ± 0.5	3.5 ± 0.2	33.4 ± 0.5	21.8 ± 1.4	1.6 ± 0.2
b (13)	9.2 ± 0.2	97 ± 11	10.5 ± 1.0	27.9 ± 0.5	2.4 ± 0.2	3.1 ± 0.1	26.4 ± 0.6	33.1 ± 0.8	6.0 ± 0.3
c (6)	8.2 ± 0.3	78 ± 10	9.6 ± 1.3	25.5 ± 0.9	2.4 ± 0.4	3.1 ± 0.6	24.3 ± 0.6	37.0 ± 1.6	6.3 ± 0.4
d (14)	11.4 ± 0.4	85 ± 7	7.3 ± 0.5	30.3 ± 0.6	3.4 ± 0.2	3.7 ± 0.2	32.7 ± 0.7	26.2 ± 1.2	2.2 ± 0.2

^a Number in parentheses is the number of rats per group. Group a: fed, killed at 8:30 a.m.; Group b: fasted from 8 p.m. to 8:30 a.m.; Group c: fasted from 8 p.m. to 6 p.m.; Group d: fed, killed at 8 p.m., at start of fasting period for Group b.

of the rats fasted overnight were significantly less than those of the fed overnight (Table I, groups a and b). Because of the difference in liver weights, the concentration (mg/gm) of liver phospholipid and triglyceride was significantly higher in the fasted group (Tables I and II), although the total amount of phospholipid and triglyceride did not significantly change. Continuation of fasting for 22 hours (group c) produced an additional decrease in liver weight, phospholipid, and triglyceride, but no additional changes in phospholipid and

triglyceride concentration. The changes in the rats fasted for 22 hours were similar to the changes observed previously in rats fasted for 24 hours (1).

In the rats fasted overnight (group b), the relative proportion (wt. %) of phospholipid arachidonate was significantly higher, and the wt. % of linoleate was significantly lower than in the fed rats (group a). If calculated in terms of milligrams of fatty acid per liver, these changes represented a small increase in arachidonate and a 24% decrease in linoleate (88 mg of arachidonate, group b cf. 84 mg of arachidonate group a; 25 mg of linoleate, group b cf. 33 mg of linoleate, group a).

Comparison of group a (8:30 a.m. fed) and group d (8 p.m. fed) gives an indication of diurnal fluctuations in liver lipid composition. In group d, the wt. % of phospholipid arachidonate was slightly higher and that of linoleate slightly lower.

With respect to triglyceride composition, the rats fasted overnight (group b) had significantly higher proportions of arachidonate and linoleate and significantly lower proportions of oleate, palmitate, and palmitoleate than did the fed rats at 8 a.m. (group a), although the total amount of triglyceride fatty acid per liver was not significantly changed. Fasting 22 hours caused a significant decrease in total triglyceride, with additional small decreases in the relative proportions of oleate and palmitate, together with an increase in linoleate. In the fed rats killed at 8 p.m., the proportion of triglyceride fatty acids was

quite similar to that of the 8 a.m.-fed rats, except for an increase in linoleate.

In summary, these results show that fasting for only 12 hours can produce significant changes in liver fatty acid composition. Consequently, feeding or fasting procedures should be carefully regulated in studies of liver lipid composition if comparable results are to be obtained in different experiments.

Summary. Fasting rats overnight from 8 p.m. to 8:30 a.m. produced significant changes in liver weight and liver fatty acid composition in comparison with fed rats killed at 8 p.m. and 8:30 a.m. Feeding and fasting procedures and time of sacrifice of animals should be carefully standardized in studies of liver lipid composition to minimize variations related to the eating patterns of the animals.

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Effect of Severe Hemorrhage on the Hemoglobins of a Virginian White-Tailed Deer (*Odocoileus virginianus*)* (32616)

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Since the original observation that sheep, homozygous or heterozygous for hemoglobin A (Hb-A), will produce a new Hb-type (Hb-C) during severe anemia (5,13) a similar phenomenon has been shown to occur in goats (7), but not in cattle (4). Structural studies have indicated that the two hemoglobins of

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