Daily Variation in Liver Tryptophan, Tryptophan Pyrrolase, and Tyrosine Transaminase in Rats Fed *ad Libitum* or Single Daily Meals (34530)

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Daily rhythms of tryptophan pyrrolase (1–3) and tyrosine transaminase (4–9) in liver and of tryptophan in plasma (1) and liver (10) of rats and mice fed *ad libitum* have been reported. Normally rats and mice consume most of their food at night (11) when fed *ad libitum,* so the earlier studies have been essentially of animals whose food intake was during the dark period of the 24-hr day. We have shown that restricting food intake of rats to a 4-hr segment of the light period shifts the rhythm of liver tyrosine transaminase by 12 hr (12). The control of food intake thus is a useful way of studying mechanisms of the enzyme variations.

The mechanisms accounting for the daily rhythms in tryptophan pyrrolase and in tyrosine transaminase are not established. Administration of tryptophan can elevate both enzymes, and there is some indication (13) that it acts in both cases at the level of transcription. Tryptophan can also enhance polysome aggregation (14, 15) and that effect could increase synthesis of these enzymes at the translational level. Tryptophan has been considered as the mediator of the tryptophan pyrrolase rhythm (1) and has been suggested to cause the rhythm of tyrosine transaminase (10). For these reasons, we have studied the effect of the pattern of food intake on the rhythms of tryptophan, tryptophan pyrrolase, and tyrosine transaminase in rats and now report the results.

Methods. Male albino rats, derived from the Wistar strain and obtained from a local supplier, were used. The rats weighed about 150 g each at the start of the experiment. One group of rats were given Purina Lab Chow and water *ad libitum.* The second group received water *ad libitum,* but were given food only from 8 AM to noon. For 2 weeks, all animals were kept individually in cages in a room with lights on from 7 AM to 7 PM. The rats were then killed by decapitation. Their livers were removed quickly and frozen on dry ice, then stored frozen prior to analysis. Blood was collected in heparinized tubes. The tubes were centrifuged, and aliquots of plasma were stored frozen.

Tryptophan pyrrolase (16), tyrosine transaminase (17), and tryptophan (18) were measured in liver, and corticosterone (19) in the plasma samples.

Results and Discussion. Table I shows the levels of tryptophan pyrrolase and of tryptophan in the livers of rats fed *ad libitum* or fed only a single daily meal. In rats fed *ad libitum,* tryptophan was lowest at 2 PM and rose sharply to its highest level at 8 PM. At each time studied, tryptophan in rats fed a single daily meal was lower than in rats fed *ad libitum.* The lowest value in the meal-fed rats was at 5 PM and the highest value was at 11 PM. These times are only 3 hr different from the nadir and peak in rats fed *ad libitum.* In other words, there was not much shift in the tryptophan rhythm.

In contrast, the rhythm in tryptophan pyrrolase was markedly altered by changing the pattern of food intake. In rats fed *ad libitum,* the highest value was at 8 PM and the lowest values at 11 PM and 2 PM. These results agree well with those of others (1). In meal-fed rats, the highest value was at 11 AM, and the lowest values were at 5 and 8 PM. That is, the peak of the rhythm in meal-fed rats was approximately at the nadir in *ad libitum* fed rats, and vice versa. This shift

TABLE I. Liver Tryptophan Pyrrolase and Tryptophan.[a]

Time	Tryptophan pyrrolase (mμmoles/min/g)		Tryptophan (mμmoles/g liver)	
	Ad libitum	Meal-fed	*Ad libitum*	Meal-fed
2 AM	16.3 ± 1.0	25.5 ± 1.9	239.2 ± 13.7	170.7 ± 2.7
5 AM	15.6 ± 1.4	27.7 ± 1.7	228.5 ± 11.8	137.1 ± 12.1
8 AM	10.5 ± 1.5	34.2 ± 5.3	212.4 ± 18.1	170.7 ± 4.6
11 AM	9.8 ± 0.3	39.3 ± 3.4	147.8 ± 31.7	139.8 ± 14.2
2 PM	8.6 ± 0.8	32.9 ± 2.4	143.8 ± 21.0	135.7 ± 6.5
5 PM	10.9 ± 2.2	22.0 ± 1.1	184.1 ± 21.4	107.5 ± 11.2
8 PM	25.6 ± 5.1	23.5 ± 3.2	260.7 ± 4.5	174.7 ± 11.0
11 PM	22.3 ± 4.1	27.1 ± 2.5	213.7 ± 23.1	190.7 ± 6.1

[a] There were 5 rats per group. Means and standard errors of the means are shown.

in enzyme activity produced by altered feeding pattern was nearly identical to that reported earlier for tyrosine transminase (12). In addition, tryptophan pyrrolase activity was generally much higher in meal-fed rats. The only exception was at 8 PM, when tryptophan pyrrolase was highest in rats fed *ad libitum* and low in meal-fed rats. The lower tryptophan levels in meal-fed rats may be due in part to greater metabolism of tryptophan by the higher levels of tryptophan pyrrolase in these rats.

Figure 1 shows the relation between tryptophan, tryptophan pyrrolase, and tyrosine transaminase in rats fed *ad libitum* and in rats fed a single daily meal. For convenience in comparing the phase of the rhythms, all values are expressed as a percentage of the maximum in each group. Shifting the food intake from night to day did not shift the tryptophan rhythm, but did shift the rhythms of both enzymes, tryptophan pyrrolase and tyrosine transaminase. The rhythm in plasma corticosterone was only slightly different in the two groups of animals. The tyrosine transaminase rhythm is known not to be generated by nor dependent upon the rhythm of plasma corticosterone (7–9). Rapoport *et al.* (1) have suggested that the rhythm in plasma corticosterone may play at least a permissive role in the tryptophan pyrrolase rhythm, and they reported that the enzyme rhythm was absent or greatly diminished in adrenalectomized mice.

Partly on the basis of the observed relationship in rats fed *ad libitum* between tryptophan and tyrosine transaminase in liver, Wurtman *et al.* (10) have suggested that the tyrosine transaminase rhythm may result from cyclic changes in hepatic tryptophan content. The association of tryptophan and tyrosine transaminase in rats, fed *ad libitum* may be fortuitous, since it does not occur when the food intake of rats is confined to the light period. Our results thus make it doubtful that any causal relationship exists

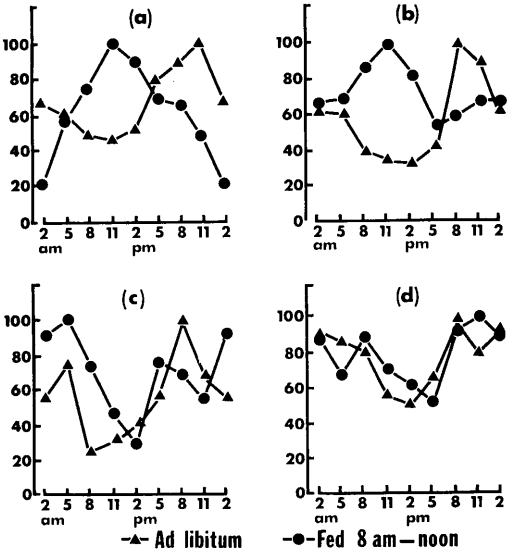

FIG. 1. Rhythm in (a) liver tyrosine transaminase, (b) liver tryptophan pyrrolase, (c) plasma corticosterone, and (d) liver tryptophan in rats fed *ad libitum* (triangles) or single daily meals (circles). Each point is the mean of determinations in five rats, and all parameters are expressed as percentage of the highest value.

between hepatic tryptophan levels and tyrosine transaminase levels.

Feigin *et al.* (20), from a study of circadian periodicity of blood amino acids in normal and adrenalectomized mice, suggested that the rhythm of tryptophan was not a likely basis for rhythmic changes in tryptophan pyrrolase. Our results would support that conclusion. Evidence that liver tryptophan pyrrolase can influence plasma tryptophan concentration has been reported (21). Our study may support that idea also, inasmuch as high tryptophan pyrrolase activity in meal-fed rats was associated with low tryptophan levels. The rhythm in liver tryptophan content was apparently not solely a consequence of rhythmic change in tryptophan pyrrolase, however, for the temporal relation between tryptophan and the enzyme was different in meal-fed rats than in rats fed *ad libitum*.

Summary. Tryptophan pyrrolase activity in the liver of rats fed a single daily meal was generally higher than in rats fed *ad libitum*, and the daily rhythm in the enzyme was shifted in meal-fed rats compared to rats fed *ad libitum*. Tryptophan in liver was lower in rats fed a single meal, but the daily rhythm in the amino acid was similar to that in rats fed *ad libitum*. The daily rhythms in tryptophan pyrrolase, tryptophan, and tryosine transaminase in liver and of corticosterone in plasma were similar in rats fed *ad libitum*, all four constituents reaching maximum between 8 and 11 PM. In rats fed a single daytime meal, the enzyme rhythms were dissociated from the tryptophan rhythm and from the plasma corticosterone rhythm. It is unlikely that the tryptophan rhythm or the plasma corticosterone rhythm generates the enzyme rhythms.

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