

preparations.^{3, 4, 5} A preliminary electrophoresis experiment has shown FSH to consist mainly of one component.

Summary. The method of preparation of FSH in highly potent form is outlined. Some of the biological properties of this fraction are described.

11775

Eating Habit and Fasting Metabolism of Rats.

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The rate of fasting metabolism depends on the level of previous food intake. In order to measure the influence on the metabolic rate of any condition which lowers the appetite one must therefore reduce the food intake of controls to that of the experimental animals in order to eliminate the influence of food level. The rats whose food intake is thus reduced but whose appetite is not affected may eat their daily ration at once while their pair mates with their decreased appetite on unlimited food supply extend their food intake over the whole day.

Table I shows the feeding habit of 18 rats during 12 days. The amounts eaten during the various daily periods are expressed in percent of the daily intake. The behavior of 9 rats that had pre-

TABLE I.
Eating Habit of Rats.
Means of 9 rats.

Nutritive condition of rats	Food eaten per day per rat g	Food eaten in % of daily intake in following periods				
		4-8 p.m. %	8-12 p.m. %	0-8 a.m. %	8-12 a.m. %	0-4 p.m. %
A. Mean of 12 days:						
1. Previously on unlimited food	14.9	16	23	47	10	4
2. " " restricted "	15.2	26	25	30	15	4
B. 1st day after on day's fast:						
1. Previously on unlimited food	15	60	7	20	0	13
2. " " restricted "	24	71	8	4	4	13

³ Greep, R. O., van Dyke, H. B., and Chow, B. F., *J. Biol. Chem.*, 1940, **133**, 289.

⁴ Fevold, H. L., Lee, M., Hisaw, F. L., and Cohn, E. J., *Endocrinology*, 1940, **26**, 999.

⁵ Jensen, H., Tolksdorf, S., and Bamman, F., *J. Biol. Chem.*, 1940, **135**, 791.

viously been on a restricted ration was essentially the same as that of their controls whose food intake had never been restricted. The previously restricted rats ate, however, during the first 4 hours of unlimited food supply 17 g of food which is more than their later average food intake during an entire day and is 71% of the intake during the first day of unrestricted intake. Does such a difference in the distribution of a given daily ration during the day affect the subsequent fasting metabolism? If such an effect would be found then it would not be sufficient to equalize the daily ration between experimental animals and controls but it would be necessary also to parallel with the controls the feeding habit of the experimental animals.

Method. Six female rats (Long-Evans strain) were given 8 g per day per rat of a food mixture which is considered qualitatively complete. Three of these rats received this ration at one time, at 4 p. m. The other 3 were given their food in 5 portions of 1.6 g at 4 p. m., 8 p. m., 12 p. m., 8 a. m., and 12 noon. Four days after the start of this regime, at the age of 88 days, the fasting metabolism of the 6 rats was measured by a respiration trial. After this trial the type of feeding was reversed. Those rats that had been fed once a day were now given their food in 5 portions and the others were now fed once per day, the total food intake remaining 8 g per day. The fasting metabolism was measured again at the age of 94 days.

The details of our respiration apparatus and the standard procedure of these trials are described elsewhere.¹ At 5 p. m. on the day prior to the respiration trial the rats were brought with water but without food to an air conditioned room kept at 30°C. The next day at 8 a. m. the rats were transferred to the multiple respiratory cabinet which was also kept at 30°C. Oxygen consumption was read at half-hour intervals and CO₂ production determined at the end of the 4-hour respiration trial.

Results. Table II shows that the metabolic rate of the 6 rats is practically the same whether they have been fed their daily ration of 8 g food once per day or in 5 portions of 1.6 g each. The difference between the 2 mean results for the daily heat production per unit of the $3/4$ power of body weight in kilograms, namely 78.3 ± 2.3 and 78.2 ± 2.3 kcal/kg ^{$3/4$} has a standard deviation of ± 3.2 kcal/kg ^{$3/4$} . The difference is thus only 1/30 of its standard deviation, it would not be regarded as significant even if it were 60 times as great as it is. One reason for this small difference is

¹Kleiber, M., *Univ. Calif. Publ. Physiol.*, in press.

TABLE II.
Effect of Once- vs. 5-Times Feeding per Day on Fasting Katabolism of Rats.

	Mean age days	Mean body wt g	Mean heat production per day per kg% kcal
6 rats fed 8* g per day once	91	161	78.3 $\pm$ 2.3
Same rats fed 1.6 g per day 5 times	91	162	78.2 $\pm$ 2.3

*N.B. These rats fed *ad lib.* ate 24, 14, 15, and 14 g of the same food on the first to the 4th day respectively after the respiration trial.

probably our method of using each rat for each of the two conditions, thus eliminating the influence of individual variation between rats on the difference between the means. Our trial shows that this individual variation in the metabolic rate between different rats is much greater than a difference resulting from a change in the number of daily feedings. For the measurement of the fasting metabolism it is thus sufficient to equalize the previous daily rations between experimental and control rats without regard to their feeding habit.

11776

Studies of Phenothiazine. X. Further Observations on Oxidation of Phenothiazine.*

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Several years ago an interesting observation was made on the oxidation of phenothiazine in the dry state. At that time no practical importance was attached to the observation, and it was not reported. As will be shown later, recent developments on the use of phenothiazine as a fungicide have pointed to the importance of this observation, which has now been repeatedly confirmed, and deemed worthy of a report.

A porcelain mortar and pestle had been used for intimately mixing a small quantity of phenothiazine with the basic rat diet routinely used in this laboratory. Subsequently the same mortar was wiped out, but not washed, and used for mixing bentonite with rat diet. It was observed that after a few minutes of mixing the whole con-

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