

which is higher than $+60^{\circ}\text{C}$, has proved most suitable (e.g. cover-glass cement according to Kroenig; Merck, Darmstadt, FRG). Thus, not only is drying of the AgNO_3 -solution prevented, but also contamination of the laboratory equipment. Moreover, the differentially stained preparations can be stored at $+4^{\circ}\text{C}$ in the dark for more than 6 months without any loss in quality.

3. Simultaneous exposure of the preparations to $+56$ – 60°C and to light from a 60-W bulb. A thermostat, which is heated and illuminated by two 60-W bulbs, has proved especially well-suited for this purpose.

This uncomplicated technique for visualizing NORs has been successfully applied to various kinds of bird and mammalian chromosomes, such as those of humans, num-

erous cervidae and different rodents. With the latter, it appeared that with this one-step procedure – quite in contrast to the original procedure – staining of the C-band regions can be avoided, thus enabling us to see clearly the NORs of the mouse (figure 2).

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Persistence of body weight cycles in dormice maintained with a limited food supply

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Summary. Dormice kept under food restriction for several months continued to exhibit body weight cycles with characteristic periods of about 2 months, despite an impaired ability to attain the high body weight levels observed in ad libitum conditions. These results imply that the cycle is controlled by a mechanism whose properties are more consistent with an endogenous oscillator than with a sequence of stages, in which the relevant stages are defined by upper and lower body weight limits.

Food intake and body weight cycles of approximately 2 months duration can be observed in dormice, *Glis glis*, kept in constant conditions of lighting and high ambient temperature^{2,3}. It has been suggested that these cycles represent accelerated hibernation rhythms². The short dormouse cycles are in many ways similar to the circannual rhythms of other hibernating mammals. Although a circannual oscillator is generally thought to be the controlling mechanism underlying the long term rhythms⁴, an alternative hypothesis, which proposes a sequence of linked stages, each taking a given amount of time to complete, must be considered^{5–8}. The stages might be based on hormonal, nutritional, neural or other internal states, and it should be possible to alter the periodicity of the cycle by slowing down, speeding up or reducing certain stages⁶. Thus, high ambient temperatures leading to an elevated energy expenditure in the weight loss phase of the body weight cycle in *Glis glis* can be thought to cause a shortening of that stage⁵. In this sense, the dormouse cycle seems to fit the sequence of stages hypothesis. This paper, however, reports findings which suggest that the cycle is controlled by an internal oscillator.

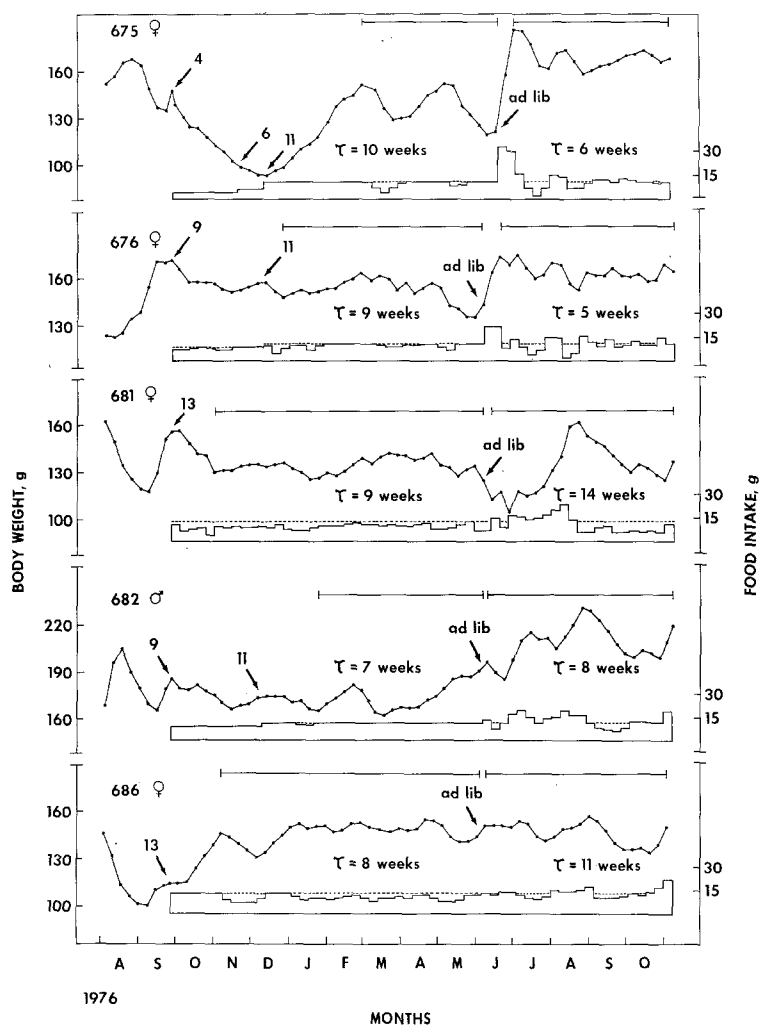
A version of the sequence of stages hypothesis – which proposes that the dormouse cycle represents oscillations between upper and lower body weight limits⁹ – predicts that dormice maintained on restricted food intake will exhibit cycles in which period is lengthened due to the greater amount of time required to achieve 'programmed' levels of fattening during the weight gain phase. On the other hand, an oscillator hypothesis predicts a preservation of periodicity during food restriction. The following experiment was carried out to test these hypotheses.

Methods. 9 dormice, purchased from a dealer in France (Stacel), were individually housed in cages $19 \times 22 \times 37$ cm, and initially provided with food (ground Purina laboratory chow, 4.25 kcal/g) and water ad libitum. The animals were kept in light/dark 12/12 (fluorescent lighting on between

08.00 and 20.00 h), and the room temperature remained $23 \pm 1^{\circ}\text{C}$. From the end of September 1976, the dormice were maintained on various levels of food restriction, the particulars of which are shown in the figure. Data from 2 animals which died and from 2 which did not cycle at any time during the experiment are not included. The daily food allowances were selected to be less than one-half the maximum daily ad lib food intake of dormice during a body weight gain phase (27 ± 1 g, $n=27$), previously determined for a group of animals kept in similar conditions. At the conclusion of the food restriction phase of the experiment, the animals were again offered food ad libitum. Throughout the restricted and subsequent ad lib period, food intakes were recorded daily and body weight was measured twice a week.

Cycle periods were estimated by periodogram analysis, using the root-mean-square excursion from mean value of each form estimate as a measure of 'amplitude'¹⁰. This method permits more objective treatment of the data than is possible with visual criteria, and greatly facilitates the analysis of 'noisy' data (e.g. animals 676 and 681). Since data from cycle phases coinciding with both the final change in food restriction level and return to ad lib were likely affected by these manipulations, they were excluded from the analysis.

Results and discussion. The body weight and food intake data, as well as cycle period estimates, are presented in the figure. The food restricted dormice continued to show body weight cycles. Further, the mean period of these cycles did not differ statistically from that obtained when food was available ad libitum (8.6 ± 0.5 [SEM] weeks and 8.8 ± 1.6 weeks, $t=0.11$, 4 df, $p>0.93$). During food restriction, periods during which the animals consumed their entire daily food ration were followed by periods of reduced food intake. It is clear that the hyperphagia which characterizes the weight gain phase of dormice in ad lib conditions is not a prerequisite to a subsequent normal weight loss phase.



Weekly body weight (dots and lines) and mean weekly food intake (histograms) of 5 dormice kept in light/dark 12/12 and $23 \pm 1^\circ\text{C}$. Arrows show when daily food allowances were changed, together with the size (g/day) of subsequent rations. Broken lines on the histograms, which represent the amounts of food offered during the restriction phase of the experiment, are carried over into the ad lib phase and permit comparison with the amounts eaten. The τ values on the left represent cycle periodicities during food restriction, while those on the right represent cycle periodicities during ad lib food intake. These estimates were obtained from periodogram analysis of the body weight data indicated by the horizontal bars.

Immediate post-restriction hyperphagia and large weight gain occurred only in those animals (675, 676) which were entering a body weight gain phase when food was again available ad libitum. The highest body weight attained during food restriction was in every case less than that observed during the period of unrestricted food availability (161.2 ± 8.4 g and 181.0 ± 12.8 g, $t = 3.21$, $p < 0.05$).

The response of these 5 animals to chronic food restriction is evidence supporting an oscillator hypothesis. The results imply that the dormouse is 'programmed' to show a particular phase of the cycle at a specified time, and that the peak and trough values actually attained are subject to constraint by factors such as rates of weight gain and loss which reflect lipogenic efficiency and locomotor activity levels. A self-

sustaining oscillator which is independent of the state of the animal's lipid reserves seems to underlie the food intake and body weight cycle in *Glis glis*, since the attainment of a high body weight is not a prerequisite to the manifestation of a weight loss phase. The considerable variability of peak and trough values both within and between animals, which is so characteristic of these cycles, also supports this interpretation.

These data, however, do not completely rule out a sequence of stages interpretation of the short dormouse cycle. It is conceivable that the relevant stages are metabolic events not directly reflected by food intake level or degree of adiposity.

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