sistent with current theories that harmonic analysis is one of the early stages of pattern recognition in humans (*5*). There are only six data points from one patient to support this theory. Nevertheless, in all cases it is clear that a given shift of the cutoff frequency is not a measure of the loss in the contrast sensitivity of other frequencies.

There are, however, difficulties before a rigorous theoretical interpretation of these findings can be attempted. The normal curve should be obtained from a larger population. Because in all patients there was some reduction in overall sensitivity, it would be worthwhile to investigate patients who have lesions not involving the visual pathways. Again it could be expected that lack of attention, or individually high threshold criteria, will produce a uniform reduction in contrast sensitivity (flat visuogram). The effect of cerebral lesions on spatial contrast sensitivity functions in humans requires further study. Besides their importance to pattern recognition theories, such studies would be revealing to the clinician who is confronted with patients that complain of difficulty with everyday visual tasks, yet have a seemingly adequate visual acuity.

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References and Notes

1. F. W. Campbell and J. G. Robson, *J. Physiol. London* **197**, 551 (1968); F. W. Campbell and L. Maffei, *ibid.* **203**, 237 (1969).
2. O. H. Schade, *J. Opt. Soc. Amer.* **46**, 721 (1956). The contrast of the grating pattern is customarily expressed as the ratio of the difference of maximum and minimum luminance over their sum. Sensitivity is measured by the inverse threshold contrast as a function of the spatial frequency of the grating. Spatial frequency is an equivalent measure of bar size in a grating pattern; that is, it is the number of alternating pairs of dark and bright bands of the pattern subtended in degree of visual angle at the observer's eye.
3. Cr. Enroth-Cugell and J. G. Robson, *J. Physiol. London* **187**, 517 (1966); F. W. Campbell, G. F. Cooper, Cr. Enroth-Cugell, *ibid.* **203**, 223 (1969).
4. F. W. Campbell and D. G. Green, *ibid.* **181**, 576 (1965).
5. C. Blakemore and F. W. Campbell, *ibid.* **203**, 237 (1969); D. A. Pollen, J. R. Lee, J. H. Taylor, *Science* **173**, 74 (1971).
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6 December 1971; revised 17 May 1972

Regulation of Testis Function in Golden Hamsters: A Circadian Clock Measures Photoperiodic Time

Abstract. *The photoperiodic testicular response of adult golden hamsters was examined by the use of a 6-hour light period coupled with dark periods of 18, 30, 42, and 54 hours. Cycle lengths of 24 and 48 hours resulted in testicular regression, whereas testicular weight was maintained by cycle lengths of 36 and 60 hours. Our data demonstrate a circadian rhythm of sensitivity to the effects of light on the photoperiodic testicular response of the hamster. The position of light relative to the circadian system (as measured by the locomotor rhythm) is critical in the response.*

Synchronization of reproduction with environmental periodicities in the field has been observed in many mammalian species (*1*). However, the regulating role of light in timing annual reproductive cycles has been demonstrated experimentally in only a few mammals. The role of photoperiod in the timing of estrus has been examined (*1, 2*), whereas few studies have considered photoperiodic control of reproductive function in males. In autumn breeders (goat and ram), short days are necessary for induction and maintenance of spermatogenesis (*3*), whereas in spring breeders such as the vole, snowshoe hare, and ferret, testicular function is stimulated by long photoperiods (*1, 2, 4*).

Hoffman, Reiter, and colleagues (*5*) demonstrated that photoperiods of LD 16 : 8 (16 hours of light and 8 hours of dark per day) or LD 14 : 10 maintained testicular size and function in the golden hamster, whereas photoperiods of LD 2 : 22 or LD 1 : 23 induced testicular regression. Gaston and Menaker (*6*) subjected male hamsters to photoperiods with 0 to 24 hours of light per day; at least 12.5 hours of light per day were required to maintain spermatogenesis and prevent testicular regression. Although light has important effects on reproductive function in at least some mammals, the mechanism by which light synchronizes breeding cycles with the environment remains unknown. In the case of photoperiodic effects, such synchronization must surely depend upon the ability of the organism to distinguish one naturally occurring day length from another. This implies the participation of a biological time-measuring system in the reproductive response to light.

The hypothesis that an endogenous circadian clock mediates the measurement of photoperiodic time was advanced by Bünning to explain the mechanism of photoperiodic time measurement in plants (*7*). Bünning proposed that the organism possesses an endogenous circadian (approximately 24-hour) rhythm of sensitivity to photoperiodic induction by light. In Bünning's model, photoperiodic induction by long days occurs when light extends into the photoinducible phase of the rhythm. On short days, induction fails because light is restricted to the nonsensitive phase of the rhythm (*8*). A major difficulty in executing direct and critical tests of Bünning's hypothesis derives from the prediction that light will have a dual role in photoperiodic systems; in addition to its action as inducer of the photoperiodic response, light will also act as a "Zeitgeber" that entrains (synchronizes) the photoperiodic sensitivity rhythm through its action on the circadian clock to which that rhythm is coupled (*9*). Thus any critical test of the Bünning hypothesis must take into account the effect of light upon the phase of the rhythm as well as its more direct effect on the photoperiodic response (such as reproductive state). Ideally, one would like to assay the phase of the photoperiodic sensitivity rhythm during exposure to various inductive and noninductive light cycles, but the sensitivity rhythm by its nature cannot be assayed independently of induction. In testing the hypothesis it therefore becomes necessary to assume that the sensitivity rhythm is controlled by the same clock that controls another readily measurable overt circadian rhythm (such as locomotor activity) and to use that rhythm as an indicator. This requires the additional assumption that the measured rhythm bears a constant phase relation to the sensitivity rhythm. If these assumptions are made, the behavior of the overt rhythm can be taken to reflect the behavior of the hypothesized rhythm of photoperiodic sensitivity.

A circadian rhythm of sensitivity to

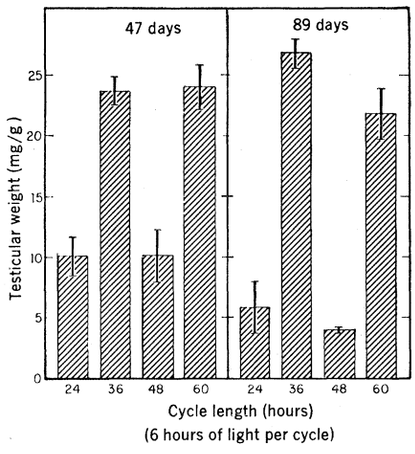

Fig. 1. Response of hamster testes to the experimental light cycles after 47 and 89 days of treatment. Height of histogram indicates group mean, and vertical bars designate standard error of the mean. Differences between groups with small testes and those with large testes are highly significant ($P < .05$, Student's t-test) in all comparisons.

photoperiodic stimuli has been shown to effect photoperiodic time measurement in some species of plants (*10, 11*), insects (*12*), and birds (*13*). No such study in a mammal has been reported. Further, an overt rhythm has seldom been measured concurrently with an assay of photoperiodic effect (*14*), and as a result little is known concerning the way in which the rhythm of photoperiodic sensitivity interacts with light cycles to produce photoperiodic effects in any species.

The male golden hamster appeared an ideal mammal in which to test Bünning's hypothesis; not only does the reproductive system respond photoperiodically (*5, 6*) but the circadian rhythm of locomotor activity is precise, stable, and extensively studied (*15*). Furthermore, increased understanding of the mechanisms involved in the control of mammalian reproduction is of considerable practical interest.

Male golden hamsters were maintained on a daily photoperiod of LD 14 : 10 for 17 weeks before the experiment. During this time the animals were transferred to light-controlled boxes and divided into four groups. Two hamsters from each group were housed in individual cages with running wheels and kept in separate boxes; their activity rhythms were recorded continuously throughout the experiment (*16*). During the experiment each group was kept on a different photoperiod, consisting of 6 hours of light coupled with a dark period to generate cycle lengths of 24 hours (LD 6 : 18), 36 hours (LD 6 : 30), 48 hours (LD 6 : 42), and 60 hours (LD 6 : 54) (*17, 18*). For each group the onset of the 6-hour light period on day 1 of the experiment coincided in real time with the onset of light in the previous LD 14 : 10 regime. Seven animals in each group were killed on day 47; the remaining 26 animals, including those in the running wheel cages, were killed on day 89.

The testicular response of the hamsters to the four experimental light cycles is illustrated in Fig. 1. The relation of testicular weight to the length of the light cycle is striking. Under photoperiods of LD 6 : 18 or LD 6 : 42, regression of testes was marked by 47 days and complete by 89 days. In contrast, animals exposed to cycles of 36 and 60 hours maintained large testes throughout the 89 days of treatment.

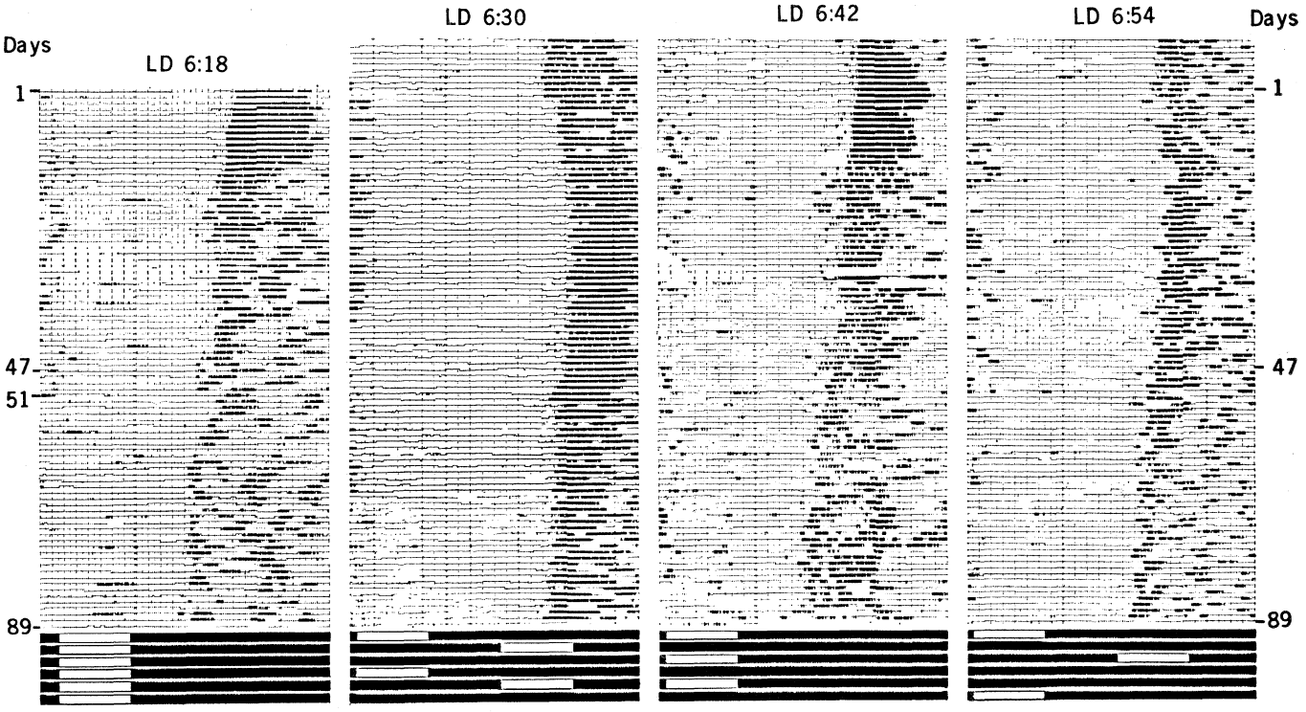

Fig. 2. Continuous wheel-running records of four hamsters, one from each experimental group. The light cycle is diagramed at the bottom of each record on a 24-hour time scale (solid black, darkness; white, fluorescent light). Each revolution of the running wheel produced a single deflection on one channel of an Esterline Angus event recorder. Each line is 1 day's record (0600 to 0600); each record is pasted under that of the preceding day. The solid areas of the activity record indicate almost continuous wheel-running, and the intermittent pen strokes represent less vigorous use of the wheel. Activity recording in three groups (36-, 48-, and 60-hour cycles) began before the start of the experiment (on day 1). Consequently the first 8 days of the record show the behavior of the rhythm in LD 14 : 10 (0830 to 2230). The gradual drift in phase of the activity rhythms of these animals reflects their synchronization to the gradual drift in phase of the 36-, 48-, and 60-hour cycles relative to real time (*18*) and does not indicate "free-running" of the circadian rhythms. On day 51 the LD 6 : 18 light cycle was advanced 1 hour (to the phase shown) to approximate the gradual drift in phase that occurred in the other light cycles.

Testicular weight is normally maintained only by photoperiods of 12.5 hours or more (*6*), yet the hamsters subjected to the 36-hour or 60-hour cycles received only one 6-hour light period every 1½ or 2½ days, respectively. Thus, whatever the concrete physiological system that measures photoperiodic time in the hamster, it does not measure the absolute duration of light, the absolute duration of darkness, or the ratio of light to darkness. In our judgment, these data can be reasonably interpreted only with reference to Bünning's hypothesis. The effect of light on the hamster's reproductive system depends primarily on the relation of the light period to the phase of the circadian rhythm of sensitivity. In the 24- and 48-hour cycles, light is present only during the insensitive phase of this rhythm and yields a "short-day" response; in the 36- and 60-hour cycles it is present during the sensitive phase of the rhythm and so maintains testicular weight (Fig. 1).

This interpretation gains support from the locomotor activity data (Fig. 2). We can arbitrarily divide the hamster's day into an active phase (beginning with the onset of activity and ending 12 hours later) and an inactive phase (the time remaining until activity onset the next day). In those light cycles allowing testicular regression (24- and 48-hour cycles), only the inactive phase receives light. In those light cycles (36- and 60-hour cycles) that maintain testis weight, light is present in both the early and late portions of the active phase. We cannot determine from these data the duration of the photosensitive portion of the cycle; the early portion of the active phase, the late portion, or both may contribute to the photoperiodic response. To put it more generally, we do not know the form or amplitude of the circadian rhythm of photosensitivity which underlies photoperiodic time measurement. We do know that such a rhythm exists.

In view of the role of the pineal gland in mediating the effects of light on the reproductive response of hamsters (*5*), the data presented herein suggest that pineal function is regulated by a circadian rhythm of photosensitivity. Unfortunately, this suggestion will be difficult to test experimentally.

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References and Notes

1. J. Aschoff, *Stud. Gen.* **8**, 742 (1955); J. Benoit and I. Assenmacher, Eds., *La Photorégulation de la Reproduction chez les Oiseaux et les Mammifères* (Publication 172, Centre National Recherche Scientifique, Paris, 1970); D. S. Farner, *Annu. Rev. Physiol.* **23**, 71 (1961); C. Thiebault, M. Courot, L. Martinet, T. Mauleon, F. du Mesnil du Buisson, R. Ortavant, J. Pelletier, J. P. Signoret, *J. Anim. Sci.* **25** (Suppl.), 119 (1966).
2. J. Hammond, Jr., *Vitamins Hormones* **12**, 157 (1954).
3. T. H. Bissonnette, *Physiol. Zool.* **14**, 379 (1941); R. Ortavant, P. Mauleon, C. Thiebault, *Ann. N.Y. Acad. Sci.* **117**, 157 (1964).
4. G. J. Davis and R. K. Meyer, *Biol. Reprod.* **6**, 264 (1972).
5. R. A. Hoffman and R. J. Reiter, *Science* **148**, 1609 (1965); R. A. Hoffman, R. J. Hester, C. Towns, *Comp. Biochem. Physiol.* **15**, 525 (19[illegible] R. J. Reiter and S. Sorrentino, Jr., *Amer. Zool.* **10**, 247 (1970).
6. S. Gaston and M. Menaker, *Science* **158**, 925 (1967).
7. E. Bünning, *Ber. Deut. Bot. Ges.* **54**, 590 (1936).
8. ———, *Cold Spring Harbor Symp. Quant. Biol.* **25**, 249 (1960); *The Physiological Clock* (Academic Press, New York, 1964); *Photochem. Photobiol.* **9**, 219 (1969).
9. C. S. Pittendrigh, *Z. Pflanzenphysiol.* **54**, 275 (1966); ——— and D. H. Minis, *Amer. Natur.* **98**, 261 (1964); in *Biochronometry*, M. Menaker, Ed. (National Academy of Sciences, Washington, D.C., 1971), pp. 212–247. Pittendrigh has reviewed and reanalyzed some of the data in this field and is responsible for emphasizing the importance of the consideration of the phase of the sensitivity rhythm in any critical test of Bünning's hypothesis.
10. K. C. Hamner, *Cold Spring Harbor Symp. Quant. Biol.* **25**, 269 (1960).
11. ——— and A. Takimoto, *Amer. Natur.* **98**, 295 (19[illegible] W. S. Hillman, *ibid.*, p. 323.
12. D. M. Peterson and W. M. Hamner, *J. Insect Physiol.* **14**, 519 (1968); W. M. Hamner, *ibid.* **15**, 1499 (1969).
13. B. K. Follett and P. J. Sharp, *Nature* **223**, 968 (1969); W. M. Hamner, *Science* **142**, 1294 (1963).
14. D. E. Brest, T. Hoshizaki, K. C. Hamner, *Plant Physiol.* **47**, 676 (1971); R. C. Bunsow, *Cold Spring Harbor Symp. Quant. Biol.* **25**, 257 (1960); R. Halaban, *Plant Physiol.* **43**, 1884 (1968); W. M. Hamner and J. T. Enright, *J. Exp. Biol.* **46**, 43 (1967); M. Menaker, in *Circadian Clocks*, J. Aschoff, Ed. (North-Holland, Amsterdam, 1965), pp. 385–395: ——— and A. Eskin, *Science* **157**, 1182 (1967).
15. V. G. Bruce, *Cold Spring Harbor Symp. Quant. Biol.* **25**, 29 (1960); J. E. Burchard, thesis, Princeton University (1958); P. J. DeCoursey, *J. Cell Comp. Physiol.* **63**, 189 (1964); C. S. Pittendrigh, in *Circadian Clocks*, J. Aschoff, Ed. (North-Holland, Amsterdam, 1965), pp. 277–297; *Cold Spring Harbor Symp. Quant. Biol.* **25**, 159 (1960).
16. Hamsters (6 weeks old) were obtained from Lakeview Hamster Colony, Newfield, New Jersey. The animals were initially housed six to eight per cage in a room and were maintained on LD 14 : 10 at room temperature until well after sexual maturity was attained. Before the experiment began, the animals were placed two per cage, two or three cages per light-controlled box. Each box was illuminated with a clock-controlled fluorescent bulb [Ken-Rad, 4 watts (F4T5/cw)] that produced an intensity of 50 to 100 lux at the floor of each cage. On day 47 of the experiment, seven animals of each group were killed; the remainder were housed one per cage for the rest of the experiment. Food (Purina laboratory chow) and water were continually available.
17. This experimental design, one form of the "resonance" experiment, was first employed by K. C. Hamner (*10*).
18. The 36-, 48-, and 60-hour cycles were controlled by Flexopulse timers (Eagle Signal Co., Davenport, Iowa), the periods of which could be set with only limited accuracy. As a result these light cycles had period lengths approximately 1 to 3 minutes shorter than intended (for instance, the 60-hour cycle was in actuality 59 hours 57 minutes). Thus the gradual shift to the left in the phase of activity onset reflects the gradual drift to the left in the phase of the light cycle.
19. We thank S. Siddiqui and L. Mostafavi for technical assistance. Supported by NIH grants HD-03803 and GM-00836 and NIH career development award HD-9327 to M.M.

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Spontaneous Middle Ear Muscle Activity in Man: A Rapid Eye Movement Sleep Phenomenon

Abstract. *Changes in compliance of the tympanic membrane have been detected in normal human sleep, presumably due to spontaneous contraction of the stapedius and tensor tympani muscles of the middle ear. In the waking state, these muscles generally respond to loud sound (middle ear reflex). Middle ear muscle activity typically erupts before or at the onset of rapid eye movement (REM) sleep and persists throughout the REM period in a discontinuous pattern resembling that exhibited by rapid eye movements. Approximately 80 percent of all nocturnal middle ear muscle activity is contained in REM sleep. Half of the remaining 20 percent occurs in the 10-minute intervals just prior to the onset of REM sleep. Middle ear muscle activity is often associated with other phasic events such as momentary enhancement of electromyogram inhibition, apnea, and K complexes. Rapid eye movements and middle ear muscle activity, though significantly correlated in REM sleep, are not always simultaneous.*

The commencement of rapid eye movement (REM) sleep in mammals is associated with a number of distinct alterations in physiological processes. These may be categorized into two major classes, tonic and phasic (*1*). Tonic phenomena, such as desynchronization of the electroencephalogram (EEG) and active inhibition of muscle tone, are continuous throughout the course of the REM period and define its temporal limits (*2*). Phasic phenomena, which generally commence at the onset of REM sleep, are short-lived and occur intermittently until termination of the REM stage.

Phenomena that erupt episodically in REM sleep have been described (*3*).