

Rats on 22.5-Hr Light:Dark Cycles Have Vaginal Opening Earlier Than Rats on 26-Hr Light:Dark Cycles

Steven Lehrer

Veterans Administration Medical Center, Bronx, New York

Rats on 22.5-hr light:dark cycles had vaginal opening significantly earlier than rats on 26-hr light:dark cycles. This finding might explain the early menarche in blind girls: Their circadian rhythms, like those of rats on 22.5-hr cycles, might be more rapid than normal. Thus a set number of cycles can occur in a shorter time than usual, allowing puberty to take place early.

Key words: puberty, circadian rhythms, pineal

INTRODUCTION

Numerous factors play a role in the onset of puberty [Sizonenko and Aubert, 1985]. Among them are brain neurotransmitters (catecholamines and serotonin), endogenous opiate peptides, and a biological clock. Pineal melatonin secretion might also have an influence [Reiter and Ellison, 1970]. These factors initiate the activity of an oscillator in the arcuate nucleus of the hypothalamus, which proceeds to generate pulses of gonadotropin-releasing hormone (GnRH). The GnRH pulses stimulate the pituitary to release the gonadotropins, follicle-stimulating hormone (FSH) and luteinizing hormone (LH), which promote the formation of sex steroids and sexual maturation.

The daily light:dark cycle influences the functioning of the biological clock. The clock mechanism, part of which is located in the suprachiasmatic nucleus of the hypothalamus (SCN), generates many normal circadian rhythms in mammals. A previous study reported that a short light:dark cycle accelerates vaginal opening in the rat [Lehrer, 1983], a finding that suggests that the early menarche in blind girls [Zacharias and Wurtman, 1964; Magee et al., 1970] and the accelerated vaginal opening in rats reared in darkness [Relkin, 1967] are most probably caused by acceleration of normal circadian

Received January 13, 1986; accepted April 11, 1986.

Address reprint requests to Dr. Steven Lehrer, Box 114A, Veterans Administration Medical Center, 130 West Kingsbridge Road, Bronx, NY 10468.

© 1986 Alan R. Liss, Inc.

rhythms. This article reports the effect of both short and long cycles on rat vaginal opening.

MATERIALS AND METHODS

Sprague Dawley albino rats (Charles River Laboratories, North Wilmington, MA) were used in this study. Each nursing rat and her pups were housed in individual plastic cages. Animals were exposed to illumination provided by a 7.5 W incandescent bulb 40 cm above the cage. The bulb was connected to a Flexopulse HG110A6 timer (Eagle Signal Company, Davenport, IA). Food and water were provided ad lib. Animals were exposed to the light:dark cycle being studied from birth. The pups were examined for vaginal opening at varied times during the day. If the examination time did not coincide with the lights-on phase of the light:dark cycle, a dim red light from a flashlight, covered with red cellophane, provided illumination. Only data from those animals having vaginal opening on Tuesday, Wednesday, Thursday, or Friday were collected for this study.

RESULTS

Results of the study are presented in Table 1. Three different light:dark cycles were used: 1) a short cycle (11.25 hr light:11.25 hr dark; that is, a 22.5-hr cycle); 2) a "normal cycle"; (12 hr light:12 hr dark, a 24-hr cycle); and 3) a long cycle (13 hr light:13 hr dark; a 26-hr cycle).

As can be seen in Table 1, vaginal opening was earliest on the 22.5-hr cycle, next earliest on the 24-hr cycle, and latest on the 26-hr cycle. One-way analysis of variance reveals that the mean vaginal opening times of the short, normal, and long cycles were significantly different ($F_{2,64} = 4.75$; $P < 0.05$).

DISCUSSION

The results of this experiment may explain a puzzling neuroendocrine aspect of blindness, its relationship to the onset of puberty. A blind, immature rat shows only a slight delay in vaginal opening and onset of first estrus. Not being a particularly photosensitive animal, the rat is minimally affected by simple light restriction [Reiter and Ellison, 1970]. However, blinded immature female rats that have also undergone olfactory bulbectomy show a marked delay in the onset of vaginal opening and first estrus. Why anosmia should so enhance photosensitivity is still uncertain.

In light of the pineal-mediated antigonadotrophic effect associated with blindness [Reiter, 1968], it is at first difficult to understand why blindness

TABLE 1. Vaginal Opening Times in Rats in This Study (Mean \pm SD)

	22.5-hr cycle	24-hr cycle	26-hr cycle
No. of rats	21	20	26
Vaginal opening (days)	33.7 \pm 2.69	34.4 \pm 2.01	36.35 \pm 3.97

should accelerate menarche. Zacharias and Wurtman [1964] noted this phenomenon. Although their findings were later questioned by Thomas and Pizzarello [1967], a third careful study [Magee et al., 1970] confirmed that blindness can accelerate menarche. Girls with minimal or no light perception had menarche at age 12.0 years; girls with shadow vision or guiding sight experienced menarche at age 12.8 years. Moreover, in one report, rats reared in darkness had their vaginal opening significantly earlier than rats reared in an 8-hr light:16-hr dark cycle [Relkin, 1967]. Other studies, however, show that light restriction delays sexual development in rats [Reiter, 1968].

The reason for the seemingly paradoxical acceleration of puberty onset, when it occurs, is that there may be, in fact, no pineal effect at all, since the rat and the human are much less photosensitive than the hamster [Johnson and Reiter, 1978]. Puberty theoretically could be accelerated in rats reared in darkness and blind girls because of an acceleration (or shortening) of normal circadian rhythms [Lehrer, 1985].

Blind adult humans and sighted humans isolated from the normal light:dark cycle have circadian rhythms that free-run and are abnormally long, about 25 hr long [Miles et al., 1977]; however, the corresponding free-running rhythms of similarly affected children are probably shorter (higher in frequency)—perhaps 21 hr long—and lengthen with age. Such lengthening (frequency reduction) with age has been demonstrated in hamsters [Davis and Menaker, 1980].

The onset of puberty may be controlled by a biological clock that serves, in effect, as a cycle-counting mechanism [Lehrer, 1985]. When a set number of light:dark cycles has been "counted" by the clock, puberty occurs. In blind girls and the rats on short cycles described in this article, the set number of cycles will occur after a shorter time than usual, and puberty takes place early. Conversely, the set number of cycles will occur after a longer time than usual in rats on long cycles, and puberty takes place later.

The effect of blindness on menarche in girls is still controversial. Populations of blind people are heterogenous with regard to light perception; that is, some have light perception, others do not. Also, the presence or absence of light perception is not always easy to assess. However, the early menarche in blind girls certainly suggests that some type of counting mechanism is governing the onset of puberty. This notion is supported by the effects of short and long light:dark cycles on rat vaginal opening time described in this article. Further studies may delineate whether the chronobiological effect of the number of light:dark cycles is indeed a key element in sexual maturation.

LITERATURE CITED

- Davis, F.C., M. Menaker (1980) Hamster through time's window: Temporal structure of hamster locomotor rhythmicity. *Am J. Physiol.* 239:R149-R155.
- Johnson, L.V., R.J. Reiter (1978) The pineal gland and its effect on mammalian reproduction. *Prog. Reprod. Biol.* 4:116-156.
- Lehrer, S. (1983) Puberty and resonance: A hypothesis. *Mt. Sinai J. Med.* 50:39-43.
- Lehrer, S. (1985) Puberty and menopause in the human: Possible relation to gonadotropin-releasing hormone pulse frequency and the pineal gland. *Pineal Res. Rev.* 3:237-257.

- Magee, K., J. Basinska, B. Quarrington, H.C. Stancer (1970) Blindness and menarche. *Life Sci.* 9:7-12.
- Miles, L.E., M. Raynal, M.A. Wilson (1977) Blind man living in normal society has circadian rhythms of 24.9 hours. *Science* 198:421-423.
- Reiter, R.J. (1968) The pineal gland and gonadal development in male rats and hamsters. *Fertil. Steril.* 19:1009-1017.
- Reiter, R.J., N.M. Ellison (1970) Delayed puberty in blinded anosmic female rats: Role of the pineal gland. *Biol. Reprod.* 2:216-222.
- Relkin R. (1967) Pineal function relation to absolute darkness and sexual maturation. *Am. J. Physiol.* 213:99-102.
- Sizonenko, P.C., M.L. Aubert (1985) Neuroendocrine changes characteristic of sexual maturation. In: *Melatonin in Humans (Proceedings of the First International Conference on Melatonin in Humans, Vienna, Austria)*. R.J. Wurtman and F. Waldhauser, eds. Center for Brain Sciences and Metabolism, Cambridge, Massachusetts, pp. 155-178.
- Thomas, J., D. Pizzarello (1967) Blindness, biologic rhythms, and menarche. *Obstet. Gynecol.* 30:507-509.
- Zacharias, L., R. Wurtman (1964) Blindness: Its relation to age of menarche. *Science* 144:1154-1155.