Total adrenal cholesterol was determined only in the 1-minute test group with suitable controls. The test group exhibited a markedly lower total adrenal cholesterol content (14.9 percent decrease), which is significant statistically at the 5-percent level. This suggests that the adaptation response of adrenal cholesterol lags behind that of ascorbic acid in auditory stress. A recovery differential between ascorbic acid and cholesterol was also noted in the type 4 adrenal response of Sayers and Sayers (*6*).

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Role of Light in the Photoperiodic Responses of Migratory Birds

Abstract. Light-dark cycles with 12-hour and 16-hour photoperiods are known to be effective in 24-hour cycles in inducing gonadal activity and fat deposition in migratory birds. Twenty-four-hour cycles with 16-hour dark periods are not effective. To test the role of the light periods and the dark periods in a given cycle, slate-colored juncos were subjected to light in cycles which combined stimulatory photoperiods (L) and inhibitory dark periods (D) as follows: 12L–16D; 12L–20D; 16L–16D; 16L–22D; 16L–32D. The results indicate that the photoperiod, not the dark period, determined the response.

Two conspicuous changes in physiological state occur in migratory birds each spring—gonadal growth and fat deposition. In the slate-colored junco (*Junco hyemalis*), day length regulates the physiological events which result in the timing of these changes and in two separate phases of the annual cycle—the preparatory phase (previously called the refractory period) and the progressive phase. The preparatory phase occurs in the fall and requires a period of short days, with a daily dark period of 12 hours or more, for its completion. Once the preparatory phase has been completed, the progressive phase begins, in late fall and winter, and results in gonadal growth and fat deposition which reach a maximum in the spring. The rate at which testicular growth occurs and the time when fat deposits first appear are a function of the daily photoperiod. Long days, with 16-hour or 20-hour photoperiods, or continuous light, induce a rapid response. Short days, with 9-hour photoperiods, induce a response but a much slower one; a 12-hour photoperiod induces a moderately rapid response similar to that induced by photoperiods of natural days. The postulate that there are different degrees of response to the photoperiod each day which summate was advanced to explain these results (*1, 2*).

The discovery that interruption of the long night of a short day with a brief period of light results in a rate of response comparable to that of a long day (*3*) has raised the question of the role of the light and dark periods. The experiments reported here are the most recent of an extensive series which was designed to determine the role of light and darkness in both the preparatory and the progressive phases. A brief summary of the results of all of the previous experiments has been presented elsewhere (*1, 2*).

From previous studies it was known that a 12-hour photoperiod, or a daily cycle of 12L–12D, was effective in inducing the progressive phase of the gonadal cycle, but that an 8-hour or 9-hour photoperiod, or daily cycle of 8L–16D, was not (*4*). In the first experiment, therefore, a 12-hour light period, known to be effective, was alternated with a 16-hour dark period, known to be "inhibitory," to give a 28-hour cycle.

The experiment, in which 19 juncos were used, began on 11 Dec. 1956 and ended 27 May 1957. These birds had already completed the preparatory phase under natural day lengths of fall. The gonadal response is given in Tables 1 and 2. As the tables show, 16 out of the 19 individuals responded, and the testicular activity continued for several months. Data are given for each individual, because of the variation in response. This variation is typical of birds exposed to 12-hour photoperiods in a 24-hour cycle. The gonadal response simulated that observed previously in connection with a 12L–12D schedule, not only in variation in rate of response but also in duration. With 16-hour photoperiods, testicular activity ends by March 1. Seventeen of the 19 birds also showed a fat response (*5*). This occurred primarily during the period 10 January–12 February, and here again the timing was similar to that observed previously with a 12L–12D schedule (*1*). Thus, the 12L–16D schedule was found to have the same effect as a 12L–12D schedule and pointed to the duration of the light period as the effective timer of the photoperiodic response.

A weakness in the experiment was the fact that the ratio of the period of light to that of darkness was close to 1, especially if there was a carry-over effect of the photoperiod (*6*). To test further, therefore, the following schedules, with longer dark periods, were selected: 12L–20D; 16L–16D; 16L–22D; and 16L–32D. This experiment was performed in the spring, when fat deposition and gonadal

Table 1. Gonadal response in male juncos to 12L–16D cycles of light and darkness, 11 December to 27 May.

No.	Autopsy date	Vol. of testes* (mm^3)	Wt. of testes* (mg)	Stage†
252	20 Feb.	22.4	14.8	3
253	20 Feb.	1.4	1.3	1
364	20 Feb.	31.9	17.6	3
375	20 Feb.	27.1	9.6	3
264	22 Mar.	47.4	34.2	4
365	22 Mar.	9.7	7.0	3R
367	22 Mar.	142.1	110.8	5
249	15 Apr.	0.7	0.5	1
251	15 Apr.	12.1	7.2	3
267	5 May	146.5	156.6	5
366	27 May	22.3	11.0	3R
376	27 May	226.5	128.6	5
377	27 May	218.5	157.8	5

* For both testes. † Stages of testis indicate state of spermatogenesis. Stage 1 is the minimum and stage 5 is the maximum. Response is positive when stage 3 or higher is reached. The letter R following the stage indicates that regression had begun.

Table 2. Gonadal response in female juncos to 12L–16D cycles of light and darkness, 11 December to 27 May.

No.	Autopsy date	Wt. of ovary (mg)	Follicles* (mm)	Wt. of oviduct (mg)
268	19 Feb.	3.6		3.0
266	22 Mar.		0.6	5.0
250	15 Apr.	27.6	1.0	6.0
254	27 May	36.2	2.0	11.2
263	27 May	20.4	1.0	10.0
265	27 May	21.0	0.9	5.4

* Differentiated large follicles in the ovary indicate activity; hence, the diameter of the largest follicle is given when differentiated follicles are present.

growth were already under way in the natural population, but it has been shown that continuation and maintenance of these responses can be regulated by the photoperiod (*1, 2, 7*). With a schedule of 9L–15D, beginning in late April, gonadal regression to the minimum occurs within 3 weeks, and fat deposits disappear. With schedules of 12L–12D and 20L–4D, the responses continue, with a slower rate and protracted response under 12-hour photoperiods. The rate of response in 16-hour photoperiods is similar to that in 20-hour photoperiods. The experiment began on 24 April 1958 and ended on 29 May 1958. Sixty-eight juncos were used.

The results are presented in Table 3. The gonadal response continued in all groups, and complete spermatogenesis occurred. Comparison of the data for the testes of these birds with previous data in our laboratory on the growth of testes in birds exposed to photoperiods of natural day length shows that the growth rate in the two cases was similar. Of the 68 birds in this experiment, 64 lost weight and reached below-normal weights, ranging from 12 to 15 g. The loss of body weight and fat deposits was probably the result of the long periods of darkness. The continued gonadal response and the attainment of maximal gonadal size in many of the birds despite the continual loss of body weight is significant.

Table 3. Gonadal response in juncos to various cycles of light and darkness, 24 April to 29 May. For male birds (♂♂) the first number in each pair of numbers represents mean volume (in mm^3) for both testes; the range for both testes is given in parentheses. The second number represents mean weight (in mg) of both testes (range in parentheses). Numbers in brackets indicate the number of birds involved. For female birds (♀♀), the first number in each pair represents the mean ovarian weight (in mg); the second represents the mean weight of the oviduct (in mg). Ranges are given in parentheses. Four females, autopsied between 1 and 3 May, showed the following means and ranges for the ovary and oviduct, respectively: 6.9 (6.4–7.4); 4.0 (3.4–6.4).

Cycle	8–15 May	22 May	29 May
		11 Birds (♂♂)	
12L–20D	168.8 [1] (——) 150.6 (——)	217.0 [4] (194.3–276.5) 186.9 (162.8–231.4)	223.6 [6] (142.0–304.1) 208.3 (130.4–288.6)
		10 Birds (♀♀)	
12L–20D	15.3 [3] (14.0–16.2) 6.4 (2.6–10.2)	16.7 [2] (15.6–17.8) 7.8 (1.6–14.0)	18.2 [5] (13.4–22.0) 6.8 (3.2–11.8)
		3 Birds (♂♂)	
16L–16D	199.0 [1] (——) 161.6 (——)	233.0 [2] (197.7–268.2) 247.0 (211.8–282.2)	
		8 Birds (♀♀)	
16L–16D	11.7 [3] (9.2–14.6) 3.7 (2.4–5.0)	21.1 [2] (14.8–27.4) 9.6 (3.8–15.4)	16.3 [3] (10.0–20.8) 9.4 (5.4–11.6)
		6 Birds (♂♂)	
16L–22D	171.2 [2] (95.6–246.9) 121.0 (71.2–170.8)		268.4 [4] (214.1–352.6) 249.6 (193.2–312.6)
		11 Birds (♀♀)	
16L–22D	15.4 [5] (12.6–19.6) 4.8 (1.4–8.2)	15.9 [3] (14.4–17.8) 11.3 (5.4–14.6)	17.6 [3] (17.0–18.4) 8.9 (4.8–12.4)
		2 Birds (♂♂)	
16L–32D		203.4 [2] (167.5–239.3) 161.1 (138.4–183.8)	
		12 Birds (♀♀)	
16L–32D	11.0 [2] (10.0–12.0) 11.4 (10.0–12.8)	18.3 [3] (13.2–26.8) 5.1 (3.0–9.2)	19.0 [7] (12.2–24.6) 9.3 (1.4–19.8)

The results of this experiment with respect to gonads are similar to the results of experiments in which there were long daily photoperiods (12 to 20 hr/day) rather than those in which there were long daily dark periods (15 and 16 hr/day). Moreover, when the ratio of dark to light was 2:1 (16L–32D group), the response was still that induced by a long day, rather than the regression induced by a short day, such as 8L–16D, which has a similar ratio of darkness to light.

From the results of these two experiments, it appears that the photoperiod is the effective part of the daily light-dark cycle in the progressive phase of the cycle. More precisely, the reaction produced by an effective light period cannot be negated by inhibitory periods of darkness. If the reaction to light is one that involves, for example, the release of gonadotropins by the pituitary, under the control of the hypothalamus, then these gonadotropins have their stimulatory effect on the testes each day, or in each cycle, and the intervening long dark periods of each cycle do not inhibit this reaction (see *2*). A variety of effective "light treatments" has been demonstrated for the progressive phase (*1, 2*).

In the preparatory phase in the fall, on the contrary, the effective part of a 24-hour light-dark cycle is the daily dark period (*1, 2*). But in the fall, a long light period can inhibit the effectiveness of a long dark period in a 32-hour cycle (*8*).

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