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PHOTOPERIODIC CONTROL OF THE TESTICULAR CYCLE IN THE EASTERN RED-WINGED BLACKBIRD, (AGELAIUS PHOENICEUS PHOENICEUS)

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PHOTOPERIODIC CONTROL OF THE TESTICULAR CYCLE IN THE EASTERN RED-WINGED BLACKBIRD, *(A GELAIUS PHOENICEUS PHOENICEUS)*

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The annual testicular cycle of many avian species which reside in temperate zones can be divided into two stages: the recrudescent stage and the regressive stage. The recrudescent stage is characterized by an increase in testis size which reflects the transition from an aspermatogenic condition to a spermatogenic condition. At the end of the breeding season, the birds enter the regressive stage. During this period a marked decrease in testis weight and volume indicates the return to an aspermatogenic condition. This stage is maintained until the next spring when presumably the increasing photoperiod induces gonadal recrudescence. If the birds are exposed to spring-like photoperiods immediately after the beginning of the regressive stage, the testes fail to respond. This post-spermatogenic phenomenon is termed photoperiodic refractoriness.

The present investigation examines the photoperiodic regulation of the annual testicular cycle of adult Eastern Red-winged Blackbirds *(Agelaius phoeniceus phoeniceus).* The testicular cycle of captive birds maintained indoors on a natural photoperiod was compared with the recorded gonadal size of wild birds collected in Michigan between 41.5° and 43.5° latitude. In addition, studies were undertaken to determine the presence, duration, and character of the refractory period. This description of the annual testicular cycle provides background information for studies which may attempt to produce or extend the infertile period as a means of controlling the population size of this species.

Adult male birds were obtained from Fish & Wildlife Service research stations in Ohio and Maryland. Experiments were undertaken with birds kept in an indoor aviary either in individual cages on natural day lengths or photoperiod rooms. A light-dark regimen of 18 hours of light and 6 hours of dark or an LD 18:6 regimen was used in the photoperiod rooms.

Nine adult male birds were kept in the aviary on a natural photoperiod for about 13 months. These birds were divided into two groups, and the left testis width was measured on either the first or fifteenth of each month. Five birds survived the entire period of study. The remaining 4 birds were replaced before March, and data were collected from each replacement bird following a one month acclimation period.

Figure 1 illustrates the seasonal change in mean testicular width of captive and wild birds. The sample size of the wild bird data ranged from 3 to 24 animals. In March the first noticeable increase in width occurred. Maximal testicular width occurred by mid-April and remained above 6.0mm until the first part of June. The timing and level of gonadal response was strikingly similar between wild and captive birds. An apparent bimodal peak occurred between April and mid-June. This phenomenon also was noted in fluctuations of the average testis weight during the annual cycle of wild Eastern Red-winged Blackbirds collected by Brenner in Pennsylvania.

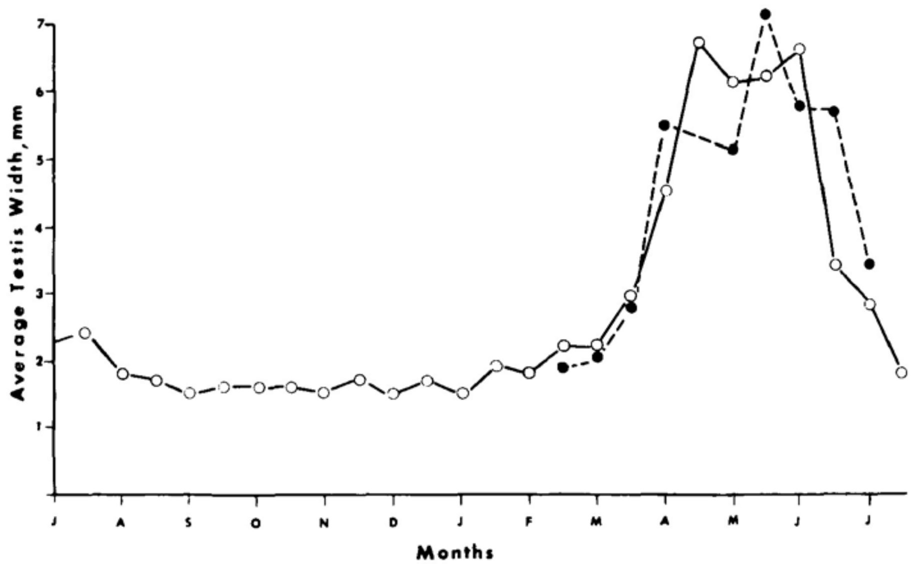

Figure 1. Seasonal change in testicular width of captive (unshaded circles) and wild (shaded circles) red-winged blackbirds.

Figure 2 is composed of a representative set of individual testicular cycles of captive birds and illustrates that each gonadal cycle had only a single peak. The bimodal peak of averaged data, therefore, may only reflect the asynchrony in the rate of gonadal development or the duration of maximal testicular width between individual gonadal cycles.

Judging from testicular size, most birds were probably fertile by early April. This inference is based on the relationship between testis size and level of spermatogenesis reported by Payne. He found that sperm were present in most non-regressing testes larger than 100 mm, which is equivalent to a left testis width of 5.2mm in birds used in this study. All captive birds possessed testis in which the width reached 5.2mm or more sometime during the spring or early summer. The average period of this maximal level response was about 56 days and ranged between 27 days to 84 days.

Testicular regression began in early June and by mid-July the testis width averaged 2.0mm. The rate of decrease in testicular width between 5.2mm and 2.0mm during regression was significantly faster than the increase in width between these limits during recrudescence.

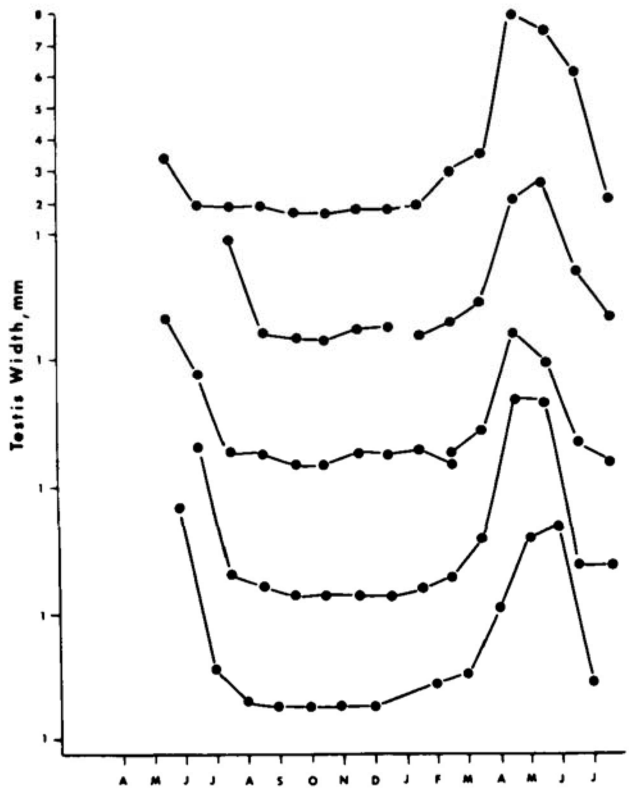

Figure 2. Representative individual testicular cycles of captive red-winged black. birds.

Figure 3 demonstrates the relationship between the annual testicular cycle, the natural photoperiod, and the occurrence of migration and nesting in Michigan birds which resided between 41.5 and 43.5° latitude. Dates of migration and nesting were prepared from records on file in the Museum of Zoology. As expected, testicular growth during March is correlated with the increase in spring day length. This relationship between the photoperiod and gonadal development continues through mid-June. By July, however, testicular regression is well advanced. Since the long day lengths which had stimulated gonadal recrudescence are now unable to support maximal gonadal activity, the birds are presumably in a state of photoperiodic refractoriness.

Payne reported that blackbirds copulate throughout the periods of nest building and egg laying up to the last egg. The period of maximal testicular width of both captive and wild blackbirds coincides with this period of copulatory activity. Spring and fall migrations, however, do not coincide with the period of maximal enlargement.

Figure 4 illustrates that the rate of regression appeared related to maximal testis size. Larger testes regressed faster in the first month after the maximal response

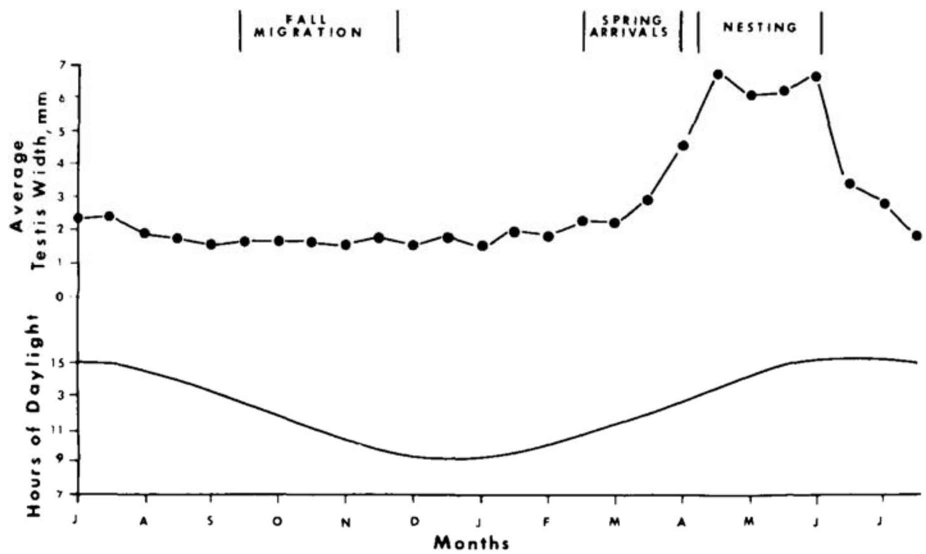

Figure 3. Annual testicular cycles relative to photoperiod in red-winged blackbirds.

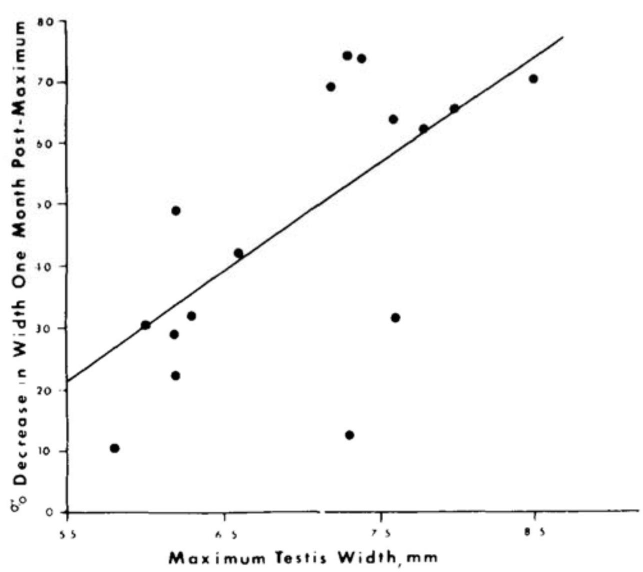

Figure 4. Relative decrease in testis size one month following maximal development in red-winged blackbirds.

than smaller testes. As a working model, it is suggested that the larger testes either produce greater amounts of a regulatory substance or they are more sensitive to a regulatory substance when compared to smaller testes.

At about monthly intervals between July, 1969 and January, 1970, birds were transferred from outdoor cages to photoperiod rooms where they were treated on a LD 18:6 regimen for about 30 days. The left testis width was measured before and after photoperiodic treatment to establish the presence and character of the refractory period. Hamner reported that the refractory period may be divided into two phases: absolute refractoriness and relative refractoriness. Absolute refractoriness may be operationally defined as a period of negative change or no change in testis size following exposure to a long-day photoperiod. Relative refractoriness, which follows immediately afterwards, is defined as a period of gradual positive change in testis size following exposure to long days. In Figure 5 the height of each bar represents the average change of testicular width, while each vertical line represents the 95% confidence limits of the mean. The average testicular width decreased slightly in groups 1 and 2, but they were not significantly different from one another or from group 3. Although the change in group 4 was significantly different from the pooled average of the first three groups, it was not significantly different from group 3 alone. The average change in group 6 was not significantly different from group 5, but it was significantly smaller than group 7.

From these data it is concluded that adult male Eastern Red-winged Blackbirds possess an absolute refractory period, which at a minimum, extends from mid-July to mid-September. (Groups 1, 2 and 3). In addition, a relative refractory period immediately follows the absolute refractory period and continues to December. (Groups 4, 5 and 6).

The mean testicular growth rate constant (k) calculated from the data collected from the birds in group 7 was .060 with a standard error of .004. Since high (k) values such as this have been shown by Farner to be characteristic when gonadal growth depends primarily upon a single environmental cue, it appears that testicular development in the Eastern Red-winged Blackbird is controlled by day length.

One group of birds, which was used in the initial analysis of the termination of the refractory period, was maintained for an additional month on the LD 18:6 regimen. It was anticipated that a second month of treatment would result in additional growth, since the testes in all but one bird had increased in size after one month of treatment. Figure 6 illustrates the average testis width of 7 birds under such a regimen. After two months of treatment, the testes of all birds had regressed. The mean testis width in mid-December was not statistically different from the mean width of untreated birds in mid-October. The mean testis width of 2.5mm in mid-November following one month of treatment, however, was different from the mean of the other two samples. The specific details of these responses are unknown. The testes may have attained a maximal level of development and then regressed so that a complete gonadal cycle was compacted within a 30 day period between mid-November and mid-December, Alternately, the testes after a small increase during the first month of treatment may have stopped growing and regressed. The data are inconclusive concerning these alternatives.

Figure 7 compares the changes in testis width following one month of a LD 18:6 regimen after termination of the absolute refractory period between

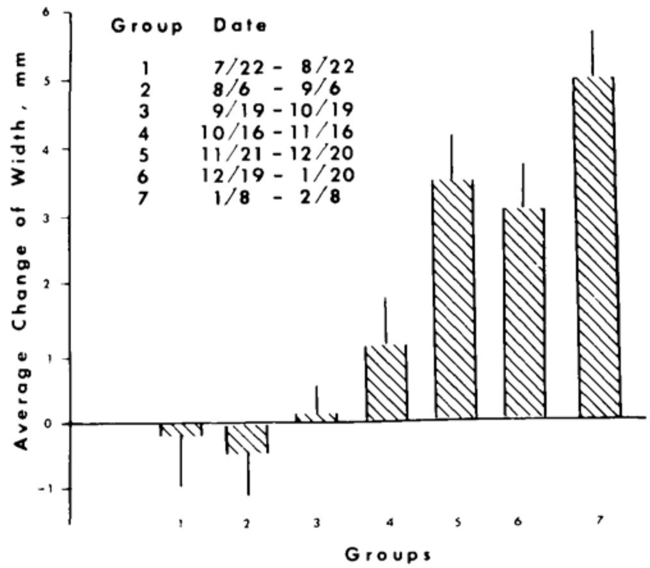

Figure 5. Average changes in testis size of red-winged blackbirds during summer and winter periods. Vertical lines represent 95% confidence limits of the means.

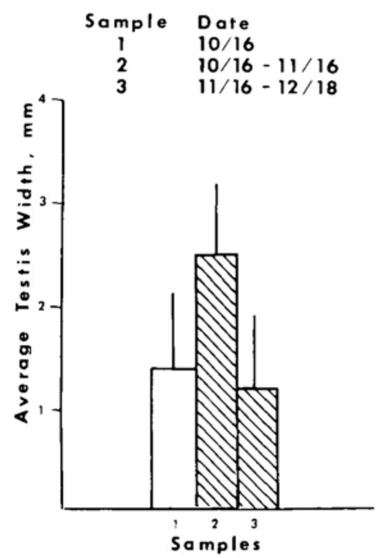

Figure 6. Testis size before (unshaded bar) and during (shaded bars) exposure of red-winged blackbirds to 18-hour days. Vertical lines represent 95% confidence limits of means.

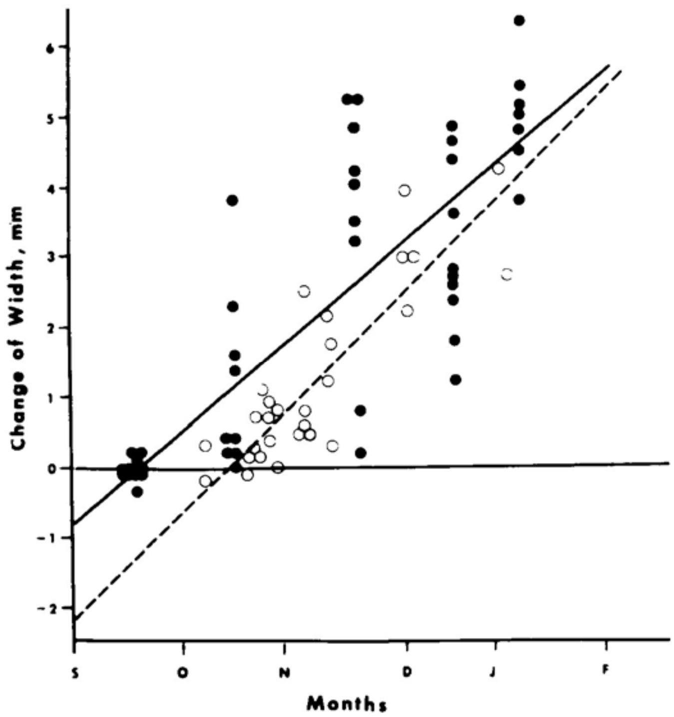

Figure 7. Change in testis size of red-winged blackbirds subjected to 18-hour days for one month after termination of the absolute refractory period. Unshaded circles represent Eastern Red-Winged Blackbirds; shaded circles, California red-winged blackbirds.

Eastern Red-winged Blackbirds and California Red-winged Blackbirds *(Agelaius phoeniceus californicus).* The regression line for the California race was calculated from raw data collected by Payne. The growth response of Eastern Red-winged Blackbird ranged from an individual minimum of 0mm on September 19 to an individual maximum of 6.4mm on January 8. Changes in testicular width of the California Red-winged Blackbird ranged from an individual minimum of -0.2mm on October 9 to an individual maximum of 4.4mm on January 4. The difference between the slopes of the two regression lines was not significant. It appears, however, that the Eastern Red-winged Blackbird responds about one month earlier. This suggests that the delay in the termination of the absolute refractory period in the California Red-winged Blackbird in turn delayed the termination of the relative refractory period. As noted earlier, the relative refractory period of the Eastern Red-winged Blackbird ends sometime between mid-December and early January. By January all of the gonadal responses fell within the range of the maximal responses observed in captive birds maintained on natural photoperiods and wild birds between mid-April and mid-June. If the same criterion is applied to the California race, the relative refractory period has not ended by early January.

In captive Eastern Red-winged Blackbirds, the period of gonadal recrudescence clearly begins in March and by early April most birds are probably fertile. Mean maximal testicular width remains above 6.0mm between mid-April and mid-June and coincides with the period of breeding activity. Gonadal growth in the absence of female birds and seasonal fluctuations in diet, rainfall, or temperature emphasizes the importance of the photoperiod as an environmental factor in the regulation of the testicular cycle.

The regressive period which begins in early June follows breeding activity, and by mid-July mean testicular width has decreased to 2.0mm. The similarity in the timing and level of gonadal responses between captive and wild birds indicates that the Eastern Red-winged Blackbird would be an excellent laboratory research animal for future investigations of the annual testicular cycle.

Under natural conditions, the regressive period is followed by the absolute refractory period. Absolute refractoriness which extends from at least mid-July to mid-September is succeeded by the relative refractory period which terminates sometime between mid-December and early January. The function of the absolute refractory period is presumably to negate the effect of the long photoperiods of summer and early autumn on spermatogenesis. The slow gonadal response to stimulatory photoperiods during the relative refractory period coupled with the progressive decrease in day length provides further protection against the occurrence of spermatogenesis in the fall.

Concurrent with this study, the biosynthesis of certain steroids from radioactive precursors is being monitored at various stages of the testicular cycle. These biochemical studies may suggest experiments in which administered steroids will interact with the photoperiod to artificially produce or extend the aspermatogenic condition characteristic of the regressive and refractory periods.

DISCUSSION

DELEGATE: Is there any information on what gonadotropin is responsible for recrudescense.

J. KERLAN: Not that I know of.

R. COON: Did you make any effort to distinguish between the Maryland birds versus the Ohio birds, or were they all put together?

J. KERLAN: They were all put together.

R. COON: Do you think there might have been or might be differences between them?

J. KERLAN: I don't know. There might be.

R. COON: Different latitude?

J. KERLAN: They are from approximately the same latitude. Taxonomists classify them as the same race.

M. KARE: At the recent meeting of the Society for the Study of Reproduction, a gentleman suggested that the removal of as few as three feathers from the top of the skull could change the reception of light in birds and alter the testicular or gonadal patterns. Is there a molt pattern related to this recrudescence in terms of absorption of light through the skull which is apparently equally or more important than that absorbed through the eye?

J. KERLAN: I don't have any data on that. Although I had the birds, you might ask why I wasn't recording moults. But that is just one of the things I didn't do. I can say that I have a feeling there is not a correlation between molt and initiation of recrudescense, but I didn't specifically study it.

D. POWER: Just very briefly relating your response to one of the other questions, it probably wouldn't have made any difference in your study, I imagine, if you had kept Maryland and Ohio birds separate; but to say that the taxonomists consider them all the same race is really not good reason for lumping them. There are subtle patterns and variations within the so-called race *Phoeniceus*, and also there really is no clear cut difference between *Phoeniceus* and the other so-called races in the Middle West. There is a gradual gradation. They are not really distinct taxa, or distinct groups, however you want to look at it.

J. KERLAN: I think that it is probably evident to everyone in the audience. The reason I used birds from two locations is that was the only way I could achieve even a small sample size. However, I am not an evolutionist, but I believe that physiologic characters are more conservative than morphologic characters. If this is the case, there may be some rationale for lumping these two groups.

T. STOCKDALE: Were all these birds taken at approximately the same time of year, Joel?

J. KERLAN: No, some of the birds were collected in the Spring and some were collected in the Fall. They were maintained in large outdoor cages for various periods of time.