

BIOENERGETICS OF LIPID DEPOSITION IN THE BOBOLINK, A TRANS-EQUATORIAL MIGRANT

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Migration places severe demands on the energy balance of a bird, especially if long distances are to be flown. Ability to store and utilize fat rapidly, that is, the ability to shift rapidly from positive to negative energy balance, is an important physiological adaptation that distinguishes migratory from nonmigratory birds (Farner, 1960). Also, equally important is the "biological clock" mechanism that synchronizes the fat deposition process with other necessary behavioral and physiological changes in such a way that timing in the annual cycle is of selective advantage to the species. Many species that breed in North America and winter in Central or South America make long nonstop flights over all or part of the Gulf of Mexico. It is especially vital in such species that maximum fat deposition be achieved at the time when birds reach the point where they must cross large bodies of water.

Recent investigations of bioenergetics of migratory birds have dealt largely with short-ranged migrants that winter in continental North America; such species become only moderately fat as compared with migrants which go to the tropics or beyond (Odum and Connell, 1956; Odum, 1960). Also, most experimental work has been concerned with the vernal migration which, in many species, is timed by increasing photoperiods of spring. The work of Farner (1955, 1960) and Wolfson (1940, 1942, 1945, 1952, 1959) on fringillids has been especially outstanding; these investigators have worked out many of the complex interrelations between photoperiod stimulation, neurohormones, *Zugunruhe*, fat deposition, and gonad enlargement. As yet few studies have dealt with long-range migrants or with the autumnal cycle that occurs on decreasing photoperiods.

The present investigation deals with bioenergetics of the autumnal fat deposition in Bobolinks (*Dolichonyx oryzivorus*), trans-equatorial migrants. A series of six experiments was carried out on captive birds and the pattern of experimentally induced fat deposition compared with that observed in wild migrants.

ACKNOWLEDGMENTS

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METHODS

Procurement and maintenance of the experimental birds.—Experimental birds were captured during spring migration in a series of 12-meter, 4-shelf Japanese mist nets set on bamboo poles between fields of oats and red clover on the experimental farms of the University of Florida at Gainesville, Florida. On May 3 and 4, 1960, 26 males and 17 females were captured in the mist nets; these birds were used in the first series of experiments that lasted from July to May. On May 3 and 4, 1961, 150 males and 20 females were netted as the flocks moved between feeding areas;

these birds provided the material for a second set of experiments and for analyses of body composition.

After removal from the mist nets, the birds were placed in large wire-cloth transportation cages divided into two sections to reduce overcrowding of any part of the cage. Water, white millet, and sessler seed were provided. During the 400-mile trip to the laboratory, the birds ate freely and often broke into song even while the vehicle was in motion. Mortality during the trip was less than one per cent. In the laboratory the birds were maintained in three large holding aviaries of $\frac{1}{2}$ -inch wire cloth, each $24 \times 30 \times 36$ inches. The birds were kept on the diet of white millet and sessler seed for about a week to ten days until weight losses normally expected in cage adaptation had been recouped. Then the diet was abruptly changed to Purina Game Bird Startena containing a minimum of crude protein (30 per cent), a minimum crude fat (3.0 per cent), minimum crude fiber (6.5 per cent) and supplemented with various amounts of minerals and required vitamins. The feed was in the form of "crumbles" with the various ingredients so finely ground and mixed that any selection by the birds was judged to be difficult. The majority of the birds accepted this new diet readily; mortality during the period of adjustment was 1.5 per cent. During the second year of this study, the manufacturer added Sulfaquinolone at the rate of 0.0175 per cent to the feed. This had no apparent effect on the birds. Also a small amount of commercial poultry Terramycin was added to the drinking water to reduce the possibility of respiratory illness, a common occurrence in caged wild birds. The Bobolinks were kept in the holding aviaries under natural daylength conditions until the start of the experiments on July 1.

Experimental design.—On July 1 groups of birds were placed under controlled daylength conditions in special cages. The cages were enclosed in four light-tight boxes constructed on $\frac{3}{4}$ -inch plywood, $30 \times 24 \times 24$ inches. Food and water consumption, weight changes, and activity rates were then measured for the next year. As shown in figure 1, an 18 inch fluorescent light which provided the main source of illumination and 10-watt "twilight" bulbs were installed. The lights were operated by a Paragon commercial poultry switch, which simultaneously turned off the main light, and turned on the "twilight" bulb for a period of 20 minutes, allowing the birds time to settle down and roost. Also a tiny neon circuit tester "tattle light" was installed in the roof of each box to simulate the amount of light present outdoors at night. This night light did not give enough illumination for photoperiodic stimulation but allowed typical nocturnal activity which Dr. William Engles (personal communication) indicates is largely or completely suppressed if the birds are retained in total darkness.

Each of the four units was equipped with a lightproof ventilating exhaust fan and louver. Within each unit, five adult birds were kept in a stainless steel Hendrix canary breeding cage, $24 \times 20 \times 22$ inches (fig. 1). These cages had removable floors and a Plexiglas partition five inches high dividing the lower half of each cage. Three hardwood perches were placed on one side of the dividing partition, and the feed dish was located on the other side. Water was provided in each case by eight 100 cc. glass tubular dispensers with narrow plastic bases which, to a large degree, retarded evaporation and prevented fouling and bath taking.

In the first series of experiments begun July 1, 1960, the birds were divided into four groups. Two of these groups, one of males and one of females, were held under simulated natural daylength; that is, a changing light regime approximating the day-light to which traveling wild migrants would be exposed, while the other two groups,

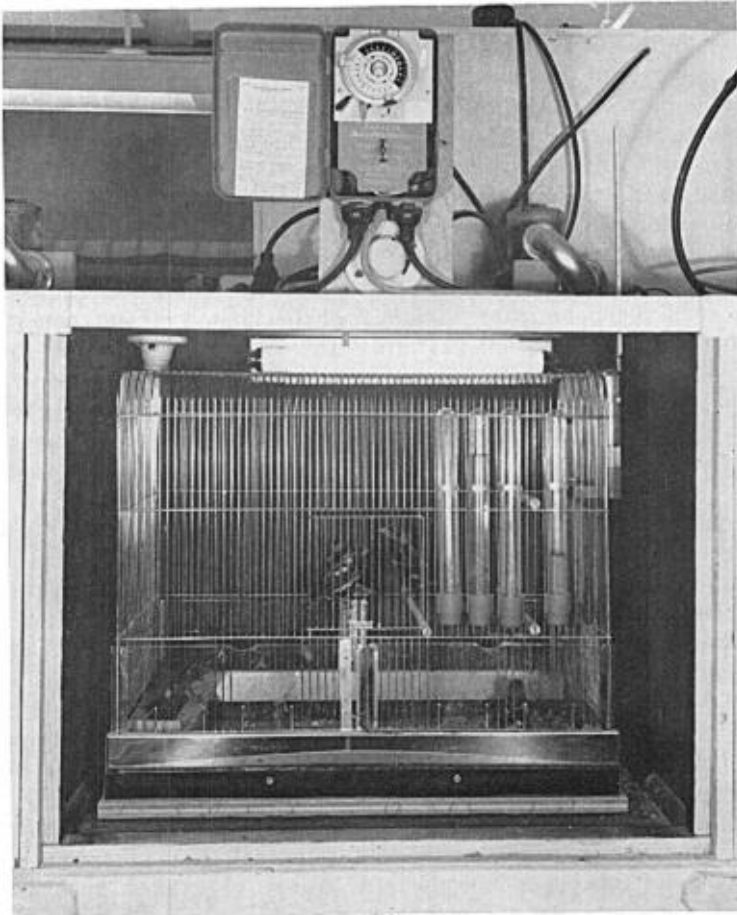


Fig. 1. A closeup of one light-tight unit showing details with inside cage and birds.

one of males and one of females, were retained under a constant 10- and 12-hour illumination, respectively. These experiments were terminated in May, 1961.

In a second series of experiments, begun in July, 1961, three groups of five male Bobolinks, selected from the birds caught in the field in May, were subjected to the following photoperiod schedules: The first group was held on simulated natural daylengths, being used as the controls. The second group was held on an accelerated schedule; that is, a schedule of very short daylengths for a period of eight weeks beginning in early July. Following this, they were returned to long photoperiods (see fig. 6 for actual photoperiods). The third group was held on a prolonged schedule; that is, the birds were kept on very long daylengths for an eight-week period beginning in July. They were then placed on short photoperiods for a three-week period (see fig. 6 for actual photoperiods). All these experiments were terminated in December, 1961.

Energy flow determination.—The method employed for measuring energy balance of the caged birds was similar to that described by Kendeigh (1949) and Siebert

(1949). Careful measurement of food intake, fecal loss, and weight changes provided the basic data for calculating the energy ingested, metabolized, catabolized, and stored as fat. At intervals of seven to 10 days uneaten food, feces, and the birds were weighed to the nearest centigram. Carefully weighed food, sufficient for the period of measurement, was supplied in a small hopper on the floor at one end of the cage, and the birds were allowed to feed *ad libitum*. Uneaten food was weighed at the same time the feces were weighed. The removable floor pan was lined with a tared piece of heavy duty aluminum freezer foil. Food and feces adhering to the birds' feet, perches, cage sides or scattered outside the cage were also collected. Feces were then meticulously separated from uneaten food by screening and subsequent hand sorting into small aluminum pans. Before final weighing, the pans containing sorted materials and the aluminum foil liner from the cage floor, to which unremovable feces adhered, were kept in a drying oven at 75 degrees C. for a period of 72 hours, the length of time previously determined as necessary to reach constant weight. As King (MS) has pointed out, it is very difficult to attain complete separation of food and feces. By careful hand picking, however, the error was reduced to an insignificant level.

The caloric energy values of both food and feces were determined in a Parr Adiabatic Oxygen Bomb Calorimeter. Both food and fecal materials, dried to a constant weight, were ground in a Willie mill to a size able to pass through a number 60 screen. Determinations for three different batches of food over the period of a year gave mean values for 10 to 20 samples each of 4.279, 4.233, 4.118 kilocalories/gm. dry weight. The mean value of 4.260 Kcal./gm. dry weight was used in the calculations in this investigation. The fecal matter, composed of intestinal egesta and nitrogenous renal excretions, had a mean value of 3.234 Kcal./gm. dry weight for 10 determinations. The caloric value of fecal matter was checked randomly throughout the duration of the experiments with no significant changes observed. No attempt was made to separate the nitrogenous excretions (ammonia, urea, and uric acid) or to determine how large a fraction of the total intestinal egesta they constituted.

For purposes of this paper "metabolized" energy, also called "available" energy (Kleiber and Dougherty, 1934), is that energy which the bird can use for work, maintenance, and storage minus the small portion lost by bacterial fermentation in the gut and as urinary waste (King and Farner, 1961; King, 1959; and Kendeigh, 1949). The approximate metabolized energy per bird per 24 hours was calculated in the following manner. First the dry weight of food consumed by the five birds used in the experiment was calculated and corrected on the basis of 10.65 per cent water content as found by oven drying of food. Total food dry weight was then multiplied by its caloric value to give the kilocalories ingested by the birds during the designated period. This figure, when divided by the number of days and the number of birds in the group, gives the gross input of Kcal./bird/day. In the same manner the caloric value of fecal output was determined. Subtracting the fecal output from food ingested gave an estimate of the metabolized energy per bird per 24 hours.

King (MS) designates the percentage of gross energy income which is assimilated by the bird as the utilization coefficient (metabolizable energy per gross energy \times 100) which in the general ecological sense can be designated as the "assimilation efficiency" (Odum, 1959).

Unless otherwise specified, several individuals (up to five) were caged and treated as a unit in the bioenergetic experiments. Therefore results expressed in cal./bird/24 hours, are calculated from the total of a specific experimental group. Bobo-

links, during both annual migration and wintering (about 70 per cent of their annual cycle), display a social behavior pattern and move in flocks of varying sizes. This social behavior must to some extent affect both the physiological activity and the psychological state of the birds. Thus, a measurement of the energy requirements of small groups or populations in captivity might provide a closer approximation to that under natural environmental conditions than would experiments employing isolated birds.

Fat determination.—Total fat extractions were made on a large number of Bobolinks collected in the field and killed at television towers during migration. Birds from the tower were picked up at dawn at Tallahassee, Florida, placed in plastic bags, and frozen by Mr. Herbert Stoddard, Sr., with fat extractions made at a later date. Fat extractions were also made from individuals kept in captivity at the end of the experimental period. Fat extraction followed the simple method developed by Odum and Connell (1956).

Measurement of Zugunruhe.—Migratory restlessness, an increase in both diurnal and nocturnal activity during migration periods, is characteristic of those species or races of birds that migrate. In the early part of this investigation a crude method of listening for movements within cages during the night was employed. By listening at the air ventilator of each cage for several hours each night, the beginning, duration, and cessation of autumnal nightly unrest could be roughly determined. The relative index of 1 through 4 was arbitrarily established to indicate estimated hops per hour as follows; 0.5 = 0–50, 1 = 51–100 hops/hr., and so on. Later, a system of perches connected to microswitches and an Esterline Angus recorder was employed, an arrangement similar to that of Farner and Mewaldt (1953). Each cage was equipped with two perches, located at each end of the cage, that were directly at-

TABLE 1
WEIGHTS OF BOBOLINKS KILLED DURING MIGRATION

Dates	Males		Females	
	Number	Mean (gm.)	Number	Mean (gm.)
Fall migration, TV tower, Tallahassee, Fla.				
Sept. 8	4	37.4	1	32.0
Sept. 11–16	12	38.6	11	33.4
Sept. 18–21	9	40.9	5	36.6
Sept. 23–24	39	42.1	30	35.3
Sept. 25–29	15	43.6	15	34.7
Sept. 30–Oct. 2	19	44.3	15	36.9
Oct. 5–8	4	49.6	7	40.2
Spring migration, TV tower, Tallahassee, Fla.				
Apr. 26–May 3	7	35.7	3	27.5
May 11–23	6	31.7	3	27.3
Spring migration, mist net capture at Gainesville, Florida				
May 5	43	31.1	22	24.7
Spring migration, mist net capture at Sapelo Island, Georgia				
May 13–15	8	26.3	8	21.2

TABLE 2
COMPARISON OF BODY COMPOSITION OF EARLY AND LATE FALL MIGRANTS KILLED
AT THE TALLAHASSEE, FLORIDA, TELEVISION TOWER¹

Dates	No.	Total wet wt. (gm.)	Water (gm.)	Fat (gm.)	Nonfat dry wt. (gm.)	Fat-free wet wt. (gm.)	Fat index ²	Water index ³
MALES								
Sept. 8-16	8	39.2 ± 1.41	18.1	12.1	8.9 ± 0.20	27.1 ± 0.82	1.36	2.03
Sept. 30-								
Oct. 8	21	45.1 ± 1.05	17.9	18.4	8.8 ± 0.18	26.7 ± 0.46	2.09	2.03
FEMALES								
Sept. 8-16	6	33.0 ± 1.23	15.8	9.7	7.5 ± 0.21	23.3 ± 0.94	1.29	2.11
Sept. 30-								
Oct. 8	11	38.3 ± 1.53	15.2	15.7	7.4 ± 0.23	22.6 ± 0.64	2.12	2.05

¹ Standard errors of means are shown for several categories.

² Fat index = grams of fat per gram of nonfat dry weight.

³ Water index = grams of water per gram of nonfat dry weight.

tached to the lever of a double-throw single-pole microswitch. By stepping on the perch, the bird discharged a capacitor which tripped another microswitch causing one channel of a 20-pen Esterline Angus operational recorder to activate and record. A relative index of activity was based on the number of strokes on the recorder charts.

RESULTS

Weight and fat deposition in wild migrants.—Data for weights of 268 wild migrant Bobolinks, arranged chronologically by season, are shown in table 1. In fall, a marked increase in weight in both males and females is evident, with late migrants averaging 8 to 12 gm. heavier than early migrants. A possible reverse trend is indicated for the spring; at least early arrivals striking the tower were heavier than late migrants captured on the ground. As would be expected, spring migrants which had presumably completed overwater journeys from the tropics were much lighter than fall migrants that had not yet made such overwater flights. Data on fat extracted from 46 of the autumnal tower-killed specimens are summarized in table 2.

TABLE 3
BODY COMPOSITION IN EXPERIMENTAL BIRDS AFTER ONE YEAR IN CAPTIVITY¹

	No.	Total wet wt. (gm.)	Water (gm.)	Fat (gm.)	Nonfat dry wt. (gm.)	Fat-free wet wt. (gm.)	Fat index	Water index
Males								
Experiments								
1 and 3	10	33.3 ± 1.21	18.5	6.5	8.5 ± 0.17	26.8 ± 1.18	0.78	2.23
Males, extra birds in holding cages —not used in experiments	11	34.1 ± 1.36	18.8	6.5	8.4 ± 0.20	27.6 ± 1.29	0.77	2.24
Females								
Experiments								
2 and 4	10	27.7 ± 1.00	15.5	5.2	7.0 ± 0.17	22.5 ± 0.74	0.74	2.21

¹ Standard errors of means are shown for several categories.

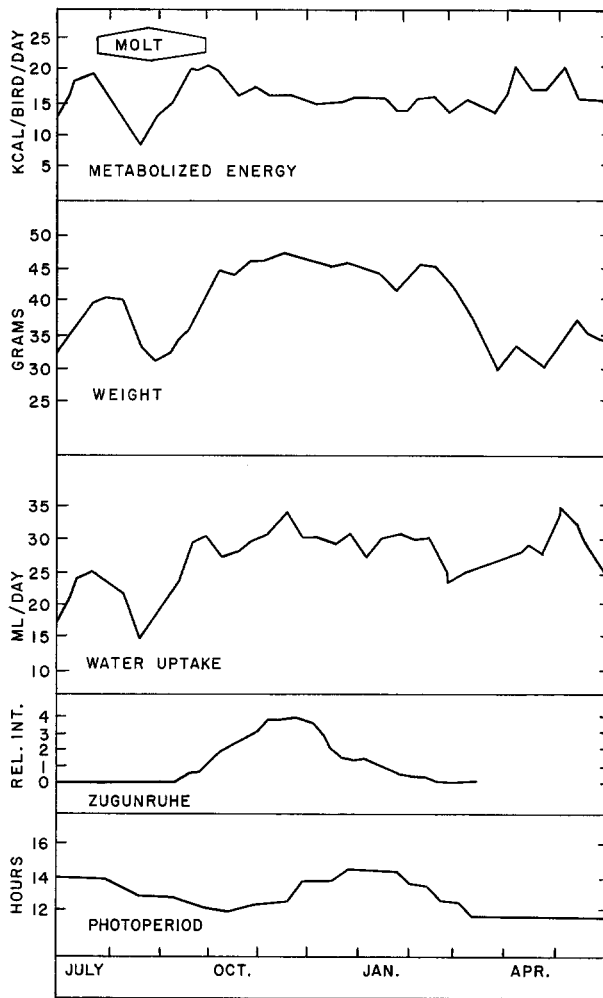


Fig. 2. Mean values for 10-day intervals of metabolized energy, body weight, water uptake and relative intensity of *Zugunruhe* in five male Bobolinks exposed to simulated natural daylength.

Since the fat-free weights of early and late migrants were essentially the same, it is clear that increase in weight of fall migrants is entirely the result of deposition of dry fat. Studies of larger series of other long-range fall migrant species also indicate that the nonfat components of the body remain essentially constant despite the large buildup and utilization of fat that occurs prior to and during migration (Odum, Rogers, and Hicks, 1964).

Body compositions of experimental birds sacrificed in May after over a year in captivity are given in table 3. These birds were somewhat heavier than wild spring migrants. However, the mean fat-free weight was similar to that of the wild fall birds although the nonfat dry weight was less. Studies of other species indicated that a slight reduction in nonfat dry weight may be expected in spring migrants

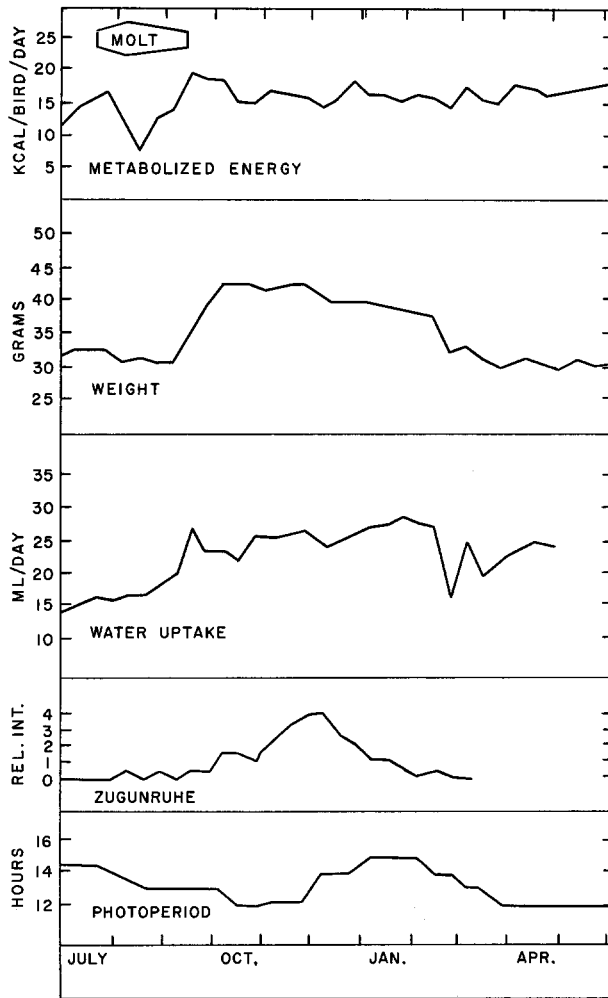


Fig. 3. Mean values for 10-day intervals of metabolized energy, body weight, water uptake and relative intensity of *Zugunruhe* in five female Bobolinks exposed to simulated natural daylength.

which have been in negative nitrogen balance during long flights (Odum, Rogers, and Hicks, 1964). It is evident that captive birds were in good condition at the end of the experiments.

First series of experiments.—The changes in metabolized energy, body weight, water consumption, and *Zugunruhe* calculated for each of the four experimental groups are shown in figures 2 to 5, together with the photoperiod regime to which each group of five birds was subjected. The annual cycle of the captive birds can be divided into five distinct periods differing in energy balance. Metabolized energy, net fat change, and estimated catabolized energy for each of the five periods are calculated in tables 4 to 7. Since, as already indicated, changes in body weight are due to changes in fat, nine Kcal./gm. was used in estimating caloric value of weight

TABLE 4
BIOENERGETICS OF MALE BOBOLINKS CAGED UNDER SIMULATED NATURAL PHOTOPERIODS

Phase of cycle	Ingested Kcal./bird/day	Metabolized Kcal./bird/day	Assimilation efficiency (per cent)	Net fat deposited (+) or utilized (-) Kcal./bird/day	Estimated catabolized energy (respiration)		<i>Zugunruhe</i> **
					Kcal./bird/day	Kcal./gm./day*	
Pre-deposition (little weight change)							
July 1-Sept. 8	25.09	15.84 ± 3.34	63	-0.03	15.86 ± 0.83	1.91 (0.59)	0.0
Rapid deposition							
Sept. 9-Oct. 18	34.60	20.23 ± 1.56	58	+2.84	17.38 ± 0.46	2.10 (0.65)	0.5-2.5
Slow utilization							
Oct. 18-Feb. 15	29.41	17.20 ± 0.57	58	+0.09	17.12 ± 0.90	2.06 (0.64)	2.5-4.0
Rapid utilization							
Feb. 16-Mar. 27	27.14	15.83 ± 0.66	58	-3.18	19.00 ± 1.92	2.29 (0.71)	0.5-0.0
Irregular response with little net weight change.							
Mar. 28-June 5	30.72	18.81 ± 1.56	58	+0.33	18.47 ± 1.49	2.23 (0.69)	No record

* Kcal. per gm. nonfat dry weight (average 8.29 gm./bird) as determined at end of experiment. Figures in parenthesis are Kcal./gm. fat-free weight (average 26.8 gm./bird).

** Range of arbitrary units.

All figures are means of five individuals; standard errors are shown for some categories.

changes. To estimate catabolized energy the estimated caloric value of weight change was added to (if weight decreased) or subtracted from (if weight increased) metabolized energy. Catabolized energy can also be considered in the broad ecological sense, "population respiration energy flow" (Odum, 1959).

As may be seen in figures 2 and 3 and tables 4 and 5, males and females responded in a similar manner to simulated natural photoperiods. Metabolized energy and weight tended to increase during July, especially in the male group, and then to decline during the peak of the fall molt in August. Late in August, before the autumnal molt was complete, ingested caloric intake increased, followed, after a lag of several days, by an increase in weight (figs. 2, 3). In figures 2 and 3, weight increase or decrease are a direct result of fat deposition. This assumption is made from data in tables 2 and 3, which clearly show that the nonfat body composition of birds remains relatively constant. During the period of hyperphagia accompanying fat deposition (September 9 to October 18) ingested caloric intake and metabolized energy averaged in both sexes about 38 per cent greater than that of the predeposition period (July 1 to September 8; tables 4 and 5).

In any type of limited growth pattern, one would expect to get a sigmoidal curve. In figures 2 and 3 the pattern of fat deposition exhibits a slow beginning, a period of very rapid deposition followed by a tapering off. The most rapid increase in weight occurred during the last week in September and the first week in October corresponding to the period of rapid weight gains in wild birds (table 1). However, in the

TABLE 5
BIOENERGETICS OF MALE BOBOLINKS CAGED UNDER SIMULATED NATURAL PHOTOPERIOD

Phase of cycle	Ingested Kcal./bird/day	Metabolized Kcal./bird/day	Assimilation efficiency (per cent)	Net fat deposited (+) or utilized (-) Kcal./bird/day	Estimated catabolized energy (respiration)		<i>Zugunruhe</i> **
					Kcal./bird/day	Kcal./gm./day*	
Pre-deposition (little weight change)							
July 1-Sept. 8	23.13	13.82 ± 2.51	60	-0.05	13.87 ± 2.48	1.92 (0.62)	0.0
Rapid deposition							
Sept. 9-Oct. 18	31.87	19.13 ± 1.95	60	+3.25	15.88 ± 0.30	2.20 (0.70)	0.5-1.5
Slow utilization							
Oct. 19-Feb. 15	28.50	16.61 ± 0.64	58	-0.37	16.98 ± 1.15	2.35 (0.75)	1.0-4.0
Rapid utilization							
Feb. 16-Mar. 27	27.41	15.92 ± 0.62	58	-1.98	17.90 ± 0.59	2.48 (0.79)	0.0-0.5
Irregular response with little net weight change.							
Mar. 28-June 5	29.57	16.94 ± 0.49	57	+0.01	16.93 ± 0.36	2.34 (0.75)	No record

* Kcal. per gm. nonfat dry weight (average 7.22 gm./bird) as determined at end of experiment. Figures in parenthesis are Kcal./gm. fat-free weight (average 22.5 gm./bird).

** Range of arbitrary units.

All figures are means of five individuals; standard errors are shown for some categories.

absence of migration captive birds remained obese all winter with only a very slow utilization of deposited fat. Finally, in late February, much of the remaining fat was utilized. This period of fat utilization, indicated by weight loss, might be explained as the result of an abortive vernal migratory phase. Premigratory deposition during the spring migration period, such as would have occurred in wild birds, failed to occur in the captives.

The response of birds on the 10-hour photoperiod (fig. 4) was similar to that of the natural daylength group, but the autumn cycle was advanced 7 to 10 days. Thus molting, hyperphagia, maximum fat weight (indicated in fig. 4 by weight) and *Zugunruhe* peaked earlier than in wild populations or the control groups (simulated natural photoperiods). The response of birds on 12-hour photoperiods was less pronounced but followed the same pattern (fig. 5). Again, as in the 10-hour group, initiation of hyperphagia and fat deposition (indicated by weight change) occurred about a week earlier than observed in the control group. Both groups of birds remained obese all winter finally decreasing in weight in early spring. Neither group held on constant short photoperiods (10 and 12 hours, respectively) exhibited any tendency toward a spring fat deposition. These birds also failed to undergo normal prenuptial (spring) molt; a few individuals exhibited an abortive molt in late March but most individuals, males and females, remained in essentially postnuptial (winter) plumage.

The efficiency of food assimilation, as shown in tables 4 to 7, is the percentage of the gross food intake that was absorbed by the gut as indicated by the ratio of

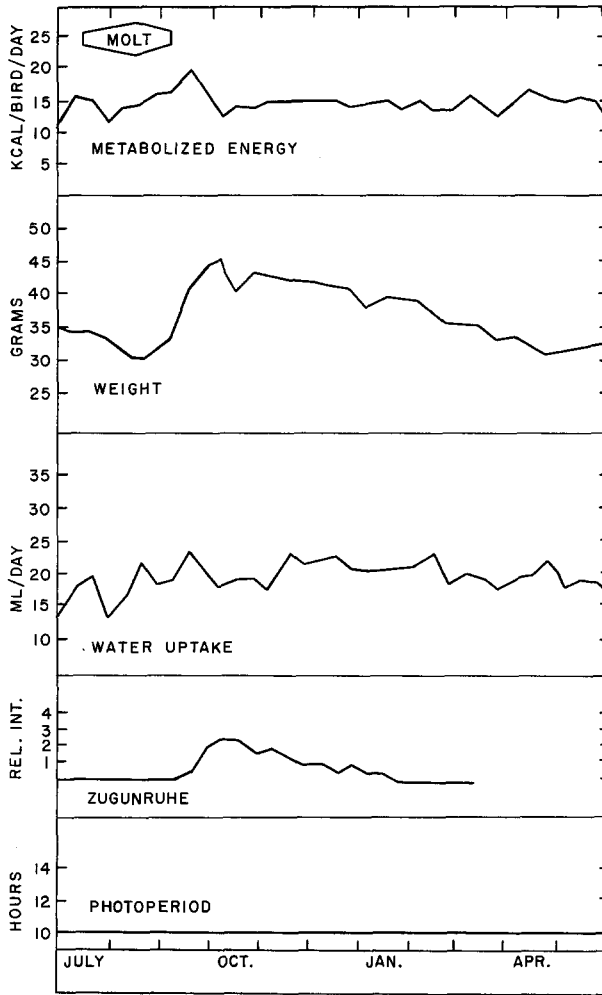


Fig. 4. Mean values for 10-day intervals of metabolized energy, body weight, water uptake and relative intensity of *Zugunruhe* in five male Bobolinks exposed to constant 10-hour daylengths.

food intake minus excretory loss divided by food intake. Calculated assimilation efficiency ranged from 57 to 63 per cent. When we consider that measurements of food consumption and excretory loss are crude at best, no consistent differences in assimilation efficiencies are evident between phases of the cycle or between groups of experimental birds.

The patterns of *Zugunruhe* as shown in figures 2 to 5 were similar in two important respects in all of the experimental groups. First, *Zugunruhe* was initiated shortly after the onset of hyperphagia and weight increase, continued to increase in intensity, and peaked after the maximum migratory fat deposition was achieved. Second, nocturnal unrest continued in the caged individuals into early winter although with a slowly declining intensity as the body weight began to decline. In experimental

TABLE 6
 BIOENERGETICS OF MALE BOBOLINKS CAGED UNDER CONSTANT 10-HOUR PHOTOPERIOD

Phase of cycle	Ingested Kcal./bird/day	Metabolized Kcal./bird/day	Assimilation efficiency (per cent)	Net fat deposited (+) or utilized (-) Kcal./bird/day	Estimated catabolized energy (respiration)		Zugunruhe**
					Kcal./bird/day	Kcal./gm./day*	
Pre-deposition (little weight change)							
July 1-Sept. 8	24.50	14.85 ± 1.24	61	-0.19	15.04 ± 0.79	1.80 (0.56)	0.0-2.5
Rapid deposition							
Sept. 9-Oct. 18	30.94	17.03 ± 2.74	55	+3.14	13.89 ± 1.29	1.67 (0.62)	0.5-2.5
Slow utilization							
Oct. 19-Feb. 15	27.81	15.85 ± 0.47	57	-0.35	16.20 ± 0.77	1.94 (0.60)	2.0-0.0
Rapid utilization							
Feb. 16-Mar. 27	26.84	15.31 ± 0.91	57	-0.99	16.31 ± 0.42	1.96 (0.61)	0.0
Irregular response with little net weight change.							
Mar. 28-June 5	27.20	15.78 ± 1.27	58	-0.15	15.93 ± 1.57	1.91 (0.70)	No record

* Kcal. per gm. nonfat dry weight (average 8.34 gm./bird) as determined at end of experiment. Figures in parentheses are Kcal./gm. fat-free weight (26.8 gm./bird).

** Range of arbitrary units.

All figures are means of five individuals; standard errors are shown for some categories.

groups 1 to 4, water consumption was measured. As shown in figures 2 to 5, water uptake increased proportionately with weight increase and fat deposition. This change cannot be due to increase in actual body water content (see water index in tables 2 and 3). This increase in water uptake might indicate a greater need of moisture for temperature regulation as the fat accumulation increases the insulation properties of the bird.

Second series of experiments: The effects of accelerated and prolonged photoperiod regimes.—Results of experiments designed to determine if timing of autumnal fat deposition could be altered by accelerated or prolonged photoperiod regimes are shown in figure 6.

In the accelerated regime of very short (8-hour) photoperiods applied in mid-summer the premolt, molting, and rapid fat deposition phases of the cycle were similar to those exhibited by birds on natural daylength but all phases occurred one to two weeks earlier. Molting was completed by September 1, at which time the mean weight was 40 gm. as compared to 32 gm. for controls. In late September when mean weight approached 45 gm. the daylength was gradually increased until a 16-hour photoperiod was reached on September 28 and held at that level for the next two months. By October 1, loss of weight and feathers became pronounced as in a typical molt. On October 3, males began to sing portions of the nuptial song. Both molting and singing continued for a period of about two weeks and then came to an abrupt halt. All birds continued to lose weight rapidly (fig. 6) and took on a very

TABLE 7
BIOENERGETICS OF FEMALE BOBOLINKS CAGED UNDER CONSTANT 12-HOUR PHOTOPERIOD

Phase of cycle	Ingested Kcal./bird/day	Metabolized Kcal./bird/day	Assimilation efficiency (per cent)	Net fat deposited (+) or utilized (-) Kcal./bird/day	Estimated catabolized energy (respiration) Kcal./bird/day	Kcal./gm./day*	Zugunruhe**
Pre-deposition (little weight change)							
July 1–Sept. 8	24.25	15.25 ± 1.31	63	+0.03	15.22 ± 1.02	1.84 (0.68)	0.0–0.5
Rapid deposition							
Sept. 9–Oct. 18	30.90	18.11 ± 0.58	59	+1.68	16.43 ± 1.42	1.91 (0.73)	1.0–3.5
Slow utilization							
Oct. 18–Feb. 15	29.24	16.63 ± 0.59	57	-0.18	16.73 ± 0.56	1.94 (0.74)	2.0–0.0
Rapid utilization							
Feb. 16–Mar. 27	29.00	16.59 ± 1.88	57	-0.73	17.32 ± 0.88	2.01 (0.77)	0.0
Irregular response with little net weight change.							
Mar. 28–June 5	28.85	16.24 ± 0.88	56	-0.55	16.79 ± 1.25	1.95 (0.75)	No record

* Kcal. per gm. nonfat dry weight (average 6.80 gm./bird) as determined at end of experiment. Figures in parentheses are Kcal./gm. fat-free weight (22.5 gm./bird).

** Range of arbitrary units.

All figures are means of five individuals; standard errors are shown for some categories.

“ratty” physical appearance. All individuals appeared to suffer some type of physiological stress, with several individuals dying in mid-December. The experiment was terminated at that time.

As can be seen in figure 6 long photoperiods (16-hour) in summer and early fall definitely greatly retarded but did not entirely prevent the fall migratory fat buildup and the fall molt. From July 21 until September 20, body weight increased slowly but no period of very rapid deposition occurred as was characteristic of birds on short daylengths. Beginning September 21, when mean weight was still less than 40 gm., the photoperiod was reduced from 16 hours to eight hours in the period of a week and held there for three weeks. After a two-week lag, a period of very rapid deposition occurred with the birds reaching a maximum body weight of almost 50 gm. about November 1 (fig. 6). In the meantime the birds had been shifted back onto long photoperiods. After another lag a rapid loss of weight occurred (fig. 6) and a new molt was initiated. This molting again stopped after about 10 days, with the birds remaining in a “patchy” plumage at termination of the experiment.

As in the first series of experiments *Zugunruhe* began at the onset of weight gain and increased in intensity as fat deposition increased. Furthermore, *Zugunruhe* decreased in proportion to weight loss under the impact of long photoperiods (fig. 6).

The effect of the autumn fat cycle on catabolized energy.—Despite the marked changes in ingested and metabolized energy that accompanied the rapid deposition and final utilization of fat, the estimated rate of catabolism per gram of nonfat

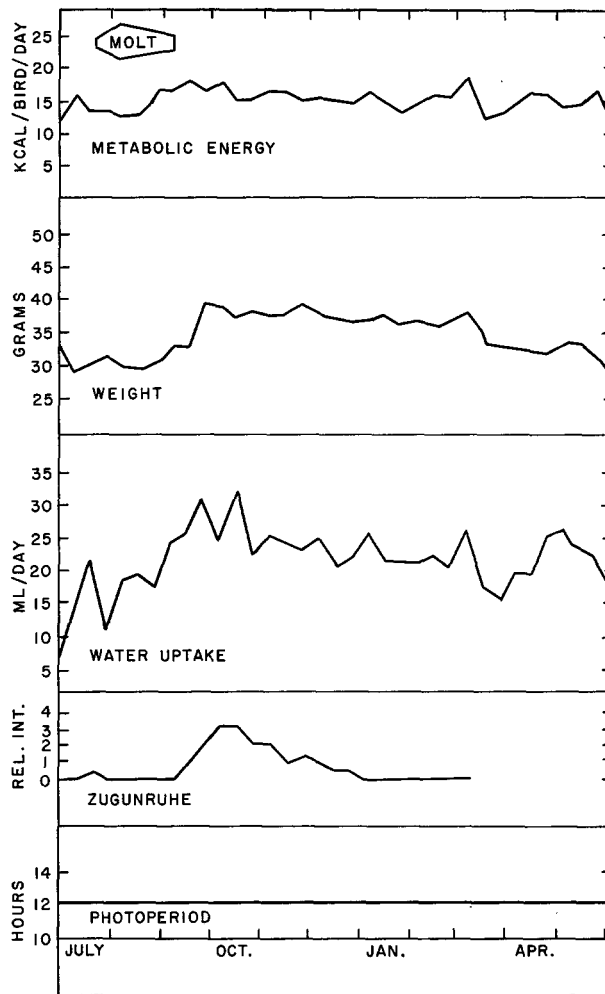


Fig. 5. Mean values for 10-day intervals of metabolized energy, body weight, water uptake and relative intensity of *Zugunruhe* in five female Bobolinks exposed to constant 12-hour daylengths.

weight remained relatively constant in all of the six experiments. However, catabolized energy did tend to average lowest in the summer predeposition period and to increase with the onset of the fat deposition cycle. Thus, the rate for groups 1 to 4 (tables 4-6) during the predeposition period was of the order of 1.9 Kcal./gm. of nonfat dry weight/day as compared with 2 to 2.2 during the period of deposition and utilization of fat. It would appear that the increased muscular activity associated with *Zugunruhe* would account for much, if not all, of this increase in catabolic rate. It was also evident that the rate of metabolism of adipose tissue itself was low even at peak deposition since very little extra catabolized energy was needed to maintain eight to 10 gm. of fat in the obese birds.

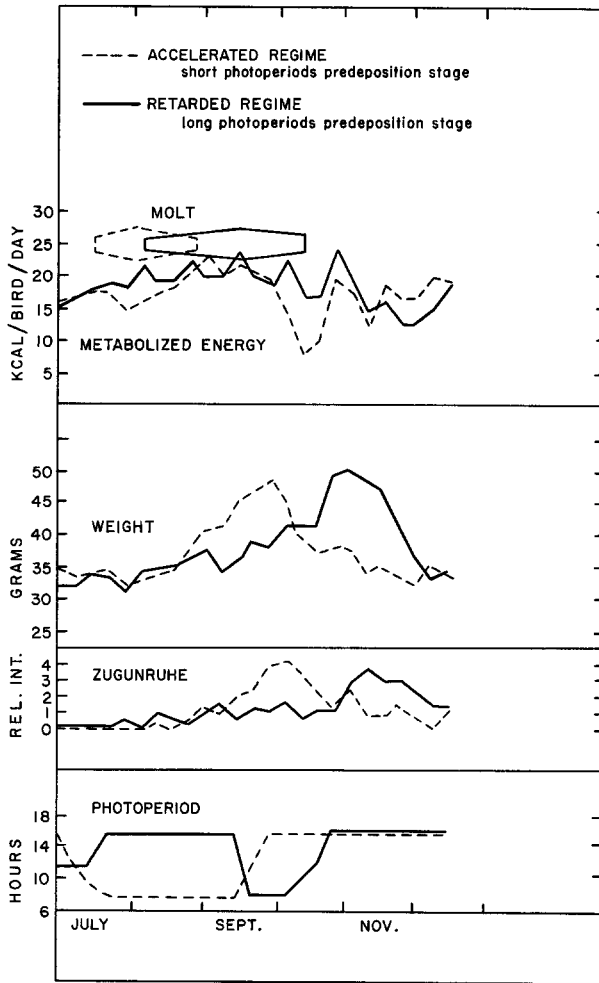


Fig. 6. Mean values for 10-day intervals of metabolized energy, body weight, water uptake and relative intensity of *Zugunruhe* in two groups of Bobolinks. One was held on an accelerated light schedule, and the other was held on a retarded light schedule.

DISCUSSION

The effect of photoperiod on the autumn fat cycle.—Weight gains during autumnal fat deposition of experimental birds subjected to the four different photoperiod regimes are compared with that of wild migrants in figure 7. Weight increases of captive birds on simulated natural photoperiods and on constant 10-hour photoperiods closely paralleled the weight gains of wild birds. Eight-hour photoperiods accelerated the response while 16 hours definitely delayed fat deposition, which, however, progressed rapidly when the daylength was reduced to eight hours. Thus, autumnal fattening occurred at least to some extent under all of the photoperiods, that is, natural, simulated natural, and constant short or long days. However, very short days (8 or 10 hours) accelerated and very long days (16 hours) retarded the response. The autumn migratory cycle in the Bobolink can thus be initiated under a wide

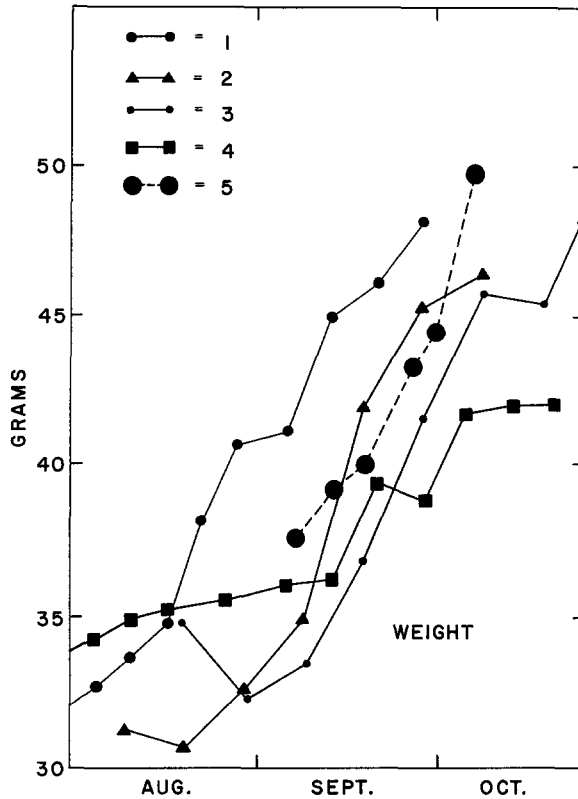


Fig. 7. Comparison of autumnal weight increase curves (minimum to maximum weight) in male Bobolinks under four experimental photoperiod regimes in comparison with weight levels of wild migrants: 1, eight-hour photoperiod, July to mid-September, then 16 hours (accelerated regime); 2, 10-hour photoperiod constant; 3, slowly decreasing photoperiod, 15 to 12 hours (simulated natural regime); 4, 16-hour photoperiod July to mid-September, then eight hours (retarded regime); 5, mean of birds killed at Gulf coast television tower during normal migratory period, September 8 to October 10.

range of photoperiods, but the decreasing daylengths of August and September are obviously important in timing the response so that the birds will reach maximum fatness at a time when long overwater flights are necessary.

It is also apparent that normal autumn molting and fat deposition can be completed in captivity by birds which had completed part of the previous spring migration but had not bred during the previous summer.

However, in the absence of actual migratory flights in the fall, the accumulated fat was not immediately utilized, and birds failed to respond normally during the spring migration period even under simulated natural photoperiods. Something must occur during migration or on the wintering grounds that resets the "biological clock." The term "biological clock" as used in this paper means an inherent internal rhythm which dictates the sequence of events, such as molting, fat deposition, migration, and so on in the annual cycle of the bird. This internal rhythm must, however, be synchronized sometime during the year with the environment to keep the

bird in phase with its physical surroundings. Marshall (1960) suggests that various types of external nervous stimuli such as special vegetation, food, the flock, and so forth, can be just as significant in the timing and regulation of the annual cycle as photoperiod.

The maintenance of migratory obesity for several months under short photoperiods (experiments 1 to 4) in contrast to the rapid loss of weight occurring when the fat birds were shifted to long photoperiods (experiments 5 and 6) was one of the most interesting results of this study. An experimental procedure involving short photoperiods to induce fat deposition followed by long photoperiods to induce utilization can provide a convenient means for the future study of lipogenesis and lipolysis in the adipose tissue itself.

The response of Bobolinks to photoperiods in autumn is, of course, just opposite from that found in the large number of studies dealing with migratory birds in the spring cycle when long rather than short photoperiods induce fat deposition. In most studies of the vernal cycle (see reviews by Farner, 1955; Wolfson, 1959) short-ranged migrants, such as sparrows in the genera *Zonotrichia* or *Junco*, will respond to long or increasing photoperiods experimentally applied at any time from December to May, but they will not respond at all if the daylength is kept at 10 hours or less.

Bioenergetics of fat deposition.—Two basic hypotheses concerning the method by which migratory species obtain the energy supply for migratory flight have been proposed. The first hypothesis, advanced by Wachs (1926), postulates that a positive energy balance is achieved during the migratory period without increased food intake by a reduction in energy expended in physiological maintenance or by an increase in the assimilation efficiency. A second hypothesis involves the establishment of a positive energy balance resulting from a direct increase in energy input (that is, hyperphagia) with the surplus energy being converted directly to fat.

Calculated assimilation efficiencies showed no consistent pattern of change in any of the groups of Bobolinks (see tables 3 to 6); certainly, there was no marked increase during the autumnal fattening period. In investigations on other species, King (1961) recorded a small increase in the assimilation efficiency for the White-crowned Sparrow (*Zonotrichia leucophrys*) during the vernal period, which seems to give support to Wach's theory. However, when this slight improvement in efficiency is converted to actual body weight, it amounts to only 0.13 gm./day. This, in itself, would hardly account for feather growth and migratory weight gains of that species. In other experiments, no cyclic changes were recorded in the assimilation efficiency so that King (*op. cit.*:137) concluded that "increase in efficiency is not a necessary concomitant of vernal fattening in the White-crowned Sparrow." It also appears that changes in assimilation efficiency are insignificant in the Bobolink and play no part in producing a positive energy balance during the fall migratory cycle.

The actual efficiency values for the captive Bobolinks ranged from 55 to 60 per cent, which are somewhat low for birds in view of the statement by King and Farner (1961:217) that assimilation efficiencies in birds range from 70 to 90 per cent, depending on the species of bird, type of food, and various other environmental factors. This low value for Bobolinks may be attributed to the artificial diet which was extremely high in protein (30 per cent).

Since the energy cost for body maintenance (that is, catabolized energy) in Bobolinks increased slightly during fat deposition, presumably as a result of *Zugunruhe*, there was no evidence that a reduction in metabolic rate played an appreciable role in providing energy for fat. While one would not want to conclude on the basis

of laboratory experiments that energy sparing mechanisms both internal and external (favorable temperature, for example) are not contributory in nature, it is clear that hyperphagia is the chief means of achieving a positive energy balance. An increase in hormonal and enzymatic activity associated with lipogenesis must, of course, accompany hyperphagia if excess metabolized energy is to be converted into fat. That the conversion is efficient is indicated by the fact that most of the increase in ingested energy could be accounted for in terms of caloric value of depot fat in so far as average balances over 10-day periods are concerned. Since hyperphagia began before the first detected increase in body weight occurred (see especially figs. 2 and 3), considerable day-to-day variations in conversion efficiency would be expected.

As King (1961) points out, the behavior of adaptive hyperphagia during the migratory cycle has not been demonstrated in wild populations. That is, no one has demonstrated in the field that birds actually eat more prior to migration. However, the weight buildup and *Zugunruhe* of captive Bobolinks so exactly paralleled weight buildup and migrations in wild birds (fig. 7) that one may conclude the hyperphagia observed in this laboratory parallels that occurring in nature.

Zugunruhe, fat levels and southward migration.—The breeding grounds of the Bobolink extend across northern United States and southern Canada between latitude 40° and 50° N and east-west from the Atlantic coast to Nebraska, Saskatchewan, and British Columbia. Small isolated breeding populations extend into Oregon. The breeding range is separated from the wintering ground of southern Brazil, Uruguay, Paraguay, and northern Argentina (between latitude 15° and 25° S) by a distance of six to seven thousand miles (see Chapman, 1890, 1909; Engels, 1959; Hamilton, 1962). The migratory flight of Bobolinks is the longest known in passerines of the Western Hemisphere. Beginning in mid-August, flocks cross the United States in southerly and easterly directions. By the time the southeasterly movement has brought birds to the Gulf coast and into the narrow Florida peninsula in September and early October, flocks may number several thousand or more individuals. From Florida, the main flight is believed to go directly south across Cuba and Jamaica and into South America where the route may follow the eastern slope of the Andes to the wintering range. The movement of the Bobolink upon reaching South America is poorly known. Chapman (1890) and Wetmore (1926) early suggested that migratory movement becomes slow with erratic wandering toward the wintering ground. Hamilton (1962) considers it likely that arrival in the southern part of the wintering grounds may not be achieved until January. The northward spring migration, mainly a reverse of the fall, presumably begins late in March. The main migratory flight reaches Florida in late April and early May.

The results of the present experimental study may contribute to a better understanding of this remarkable journey. As far as known, migratory flights begin at night; short flights may be terminated later in the same night while long flights may continue into the next day. It is important to note that *Zugunruhe*, which presumably indicates a willingness or desire to migrate at night, begins shortly after onset of the first weight increase but does not reach a peak until the maximum fat weight has been achieved. As may be seen in figures 2 to 5, *Zugunruhe* was first noted in late August and early September in captive birds when only a small amount of fat had been deposited. Thus, wild birds in August, assuming they were in the same physiological state as captives, would exhibit only a weak migratory urge and have fuel for only short flights. This would account for the slow and erratic movement of birds southeastward across the United States. The first birds reach the Gulf coast

of Florida about September 10 (see table 1 and figure 7) and are only about one-third to one-half as fat as late migrants. At this time *Zugunruhe* in captives was only about one-third the maximum intensity. Early migrants would be capable of moderately long flights, as, for example, to southern Florida or Cuba. In contrast, late migrants arriving in Florida in October exhibit maximum fatness at a time when captives are approaching a peak in *Zugunruhe*. These birds would be capable of nonstop flights from Florida to South America if we accept the estimates of Odum and Stoddard (1961).

Merkel (1937, 1938) showed a similar correlation between body weight and the intensity of *Zugunruhe* in the Garden Warbler (*Sylvia borin*) and later reported (1958) experimental data showing that in captivity the spring migratory unrest of the White-throat (*Sylvia communis*), Blackcap (*Sylvia atricapilla*) and American Robins (*Turdus migratorius*) is extended beyond the normal migratory period well into the summer months. King (1961) observed similar behavior in the White-crowned Sparrow and suggested that the duration of *Zugunruhe* is directly correlated with obesity. More recently Caldwell, Odum, and Marshall (1964) have found that autumnal migratory thrushes and warblers killed at television towers on the Florida coast are more than twice as fat as individuals of the same species killed during nocturnal migrations in Michigan. Weise (1963) further reports that the intensity of *Zugunruhe* in captives is greater in long-range than in short-range migrant species.

It would seem that the relation between the intensity of the nocturnal migratory urge and the body fat level provides a regulatory mechanism for insuring that the long flight necessary to span the Gulf of Mexico or Caribbean Sea will occur at a time when the maximum possible fuel is available.

SUMMARY

The bioenergetics of the autumnal fat deposition in the Bobolink (*Dolichonyx oryzivorus*) was investigated in a series of experiments with caged birds. Groups of five individuals, selected from birds captured the previous May during the spring migration, were exposed to controlled daylength schedules beginning in July. In the first series of experiments, running from July, 1960, to May, 1961, two groups, one of males and one of females, were held under simulated natural photoperiods, that is, a changing light regime approximating the daylength to which traveling wild Bobolinks would be exposed. Two additional groups, one male and one female, were held under a constant photoperiod of 10 and 12 hours, respectively. In the second series of experiments, running from July to December, 1961, a group of males was exposed to an 8-hour daylength until September then shifted to 16 hours. The second group of males was held under a long daylength (16 hours) then shifted to short day in September then back to a long photoperiod in October. Weight buildup accompanying autumnal migratory obesity in caged birds was compared with that observed in wild migrants accidentally killed at a Florida Gulf coast television tower.

Wild autumnal migrants in northern Florida increased in weight from a mean of 38 gm. in early September to 50 gm. in early October in males and from 32 to 40 gm. in females. The increase in weight was shown to be entirely the result of deposition of dry fat. The mean weight of spring migrants from the same locality was less than 36 gm. for males and 28 gm. for females.

The fat deposition cycle in captive birds could be divided into several phases: (1) predeposition period including the postnuptial molt, (2) period of rapid fat deposition, (3) period of obesity with a slow utilization of fat, and (4) period of rapid

utilization of fat. The period of rapid fat deposition was characterized by marked hyperphagia (38 per cent average increase in food intake). Assimilation efficiencies remained unchanged so that metabolized energy flow increased proportionally, but catabolized energy increased only slightly since most of the increased energy assimilated was stored as fat. These results indicate that hyperphagia is the chief means of achieving a positive energy balance during the autumnal premigratory fattening in the Bobolink.

Zugunruhe (nocturnal activity) began shortly after the onset of weight increase but did not reach peak intensity until maximum fat deposition was achieved.

Weight increases in birds on simulated natural and on constant 12-hour photoperiods paralleled that of wild birds in timing and magnitude. Very short photoperiods of 8 to 10 hours accelerated the autumnal fattening, while long photoperiods of 16 hours retarded it.

In the absence of actual migration, captive birds on simulated natural and short photoperiods (first series of experiments) remained obese throughout the fall and winter although they lost weight very slowly over the period of several months. In spring, the remaining fat was rapidly utilized and a partial prenuptial (spring) molt occurred, but no consistent spring premigratory fat deposition followed. Obese birds shifted to long photoperiods in the fall (second series of experiments) rapidly lost weight and began an abortive spring molt.

It is concluded that during the southward migration photoperiodism and the coupled *Zugunruhe*-migratory fat level relationship are mechanisms that synchronize maximum fat deposition, the urge to make long flights, and the seasonal arrival of migrants at geographical points where long overwater nonstop flights are necessary.

LITERATURE CITED

- Caldwell, L. D., Odum, E. P., and Marshall, S. G.
1964. Comparison of fat levels in migratory birds killed at a central Michigan and a Florida gulf coast television tower. *Wilson Bull.*, 75:428-434.
- Chapman, F. M.
1890. On the winter distribution of the bobolink (*Dolichonyx oryzivorus*) with remarks on its routes of migration. *Auk*, 7:39-45.
1909. The bobolink. *Bird Lore*, 11:137-140.
- Engels, W. L.
1959. The influence of different day lengths on the testes of a transequatorial migrant, the bobolink. *In Photoperiodism and Related Phenomena in Plants and Animals*, edited by R. B. Withrow (Publ. 55, Amer. Assoc. Adv. Sci., Washington, D.C.), pp. 759-766.
- Farner, D. S.
1955. The annual stimulus for migration: experimental and physiologic aspects. *In Recent Studies in Avian Biology*, edited by A. Wolfson (Univ. Illinois Press, Urbana, Ill.), pp. 198-237.
1960. Metabolic adaptations in migration. *Proc. XII Int. Ornith. Congr. (Helsinki)*, 1958: 196-208.
- Farner, D. S., and Mewaldt, L. R.
1953. The recording of diurnal activity patterns in caged birds. *Bird-Banding*, 24:55-65.
- Hamilton, W. J., III.
1962. Bobolink migratory pathways and their experimental analysis under night skies. *Auk*, 79:208-233.
- Kendeigh, S. C.
1949. Effect of temperature and season on the energy resources of the English sparrow. *Auk*, 66:113-127.

- King, J. R.
1961. The bioenergetics of vernal premigratory fat deposition in the white-crowned sparrow. *Condor*, 63:128-142.
- King, J. R., and Farner, D. S.
1961. Energy metabolism, thermoregulation and body temperature. *In Biology and Comparative Physiology of Birds*, edited by A. J. Marshall. Vol. II (Academic Press, New York), pp. 215-288.
- Kleiber, M., and Dougherty, J. E.
1934. The influence of environmental temperature on the utilization of food energy in baby chicks. *Jour. Gen. Physiol.*, 17:701-726.
- Marshall, A. J.
1960. Annual periodicity in the migration and reproduction of birds. *Cold Spring Harbor Symp. Quant. Biol.*, 25:499-505.
- Merkel, F. W.
1937. Zur Physiologie des Vogelzugtriebes. *Zool. Anzeiger*, 117:297-308.
1938. Zur Physiologie der Zugenruhe bei Vögeln. *Ber. Ver. Schles. Ornith.*, 23:1-72.
1958. Untersuchungen über tages- und jahreperiodische Änderungen im Energiehaushalt gekäfigter Zugvögel. *Zeits. vergl. Physiol.*, 41:154-178.
- Odum, E. P.
1959. *Fundamentals of ecology*. Second ed. (W. B. Saunders, Philadelphia).
1960. Lipid deposition in nocturnal migrant birds. *Proc. XII Int. Ornith. Congr. (Helsinki)*, 1958, 2:563-576.
- Odum, E. P., and Connell, C. E.
1956. Lipid levels in migrating birds. *Science*, 123:892-894.
- Odum, E. P., Connell, C. E., and Stoddard, H. L.
1961. Flight energy and estimated flight ranges of some migratory birds. *Auk*, 78:515-527.
- Odum, E. P., Rogers, D. T., and Hicks, D. L.
1964. Homeostasis of the nonfat components of migrating birds. *Science*, 143:1037-1039.
- Seibert, H. C.
1949. Differences between migrant and non-migrant birds in food and water intake at various temperatures and photoperiods. *Auk*, 66:128-153.
- Wachs, H.
1926. Die Wanderungen der Vögel. *Ergeb. Biol.*, 1:479-637.
- Weise, C. M.
1963. Annual physiological cycles in captive birds of differing migratory habits. *Proc. XIII Int. Ornith. Congr. (Ithaca)*, 1962, 2:983-993.
- Wetmore, A.
1926. *The migrations of birds* (Harvard Univ. Press, Cambridge).
- Wolfson, A.
1940. A preliminary report on some experiments on bird migration. *Condor*, 42:93-99.
1942. Regulation of spring migration in juncos. *Condor*, 44:237-263.
1945. The role of the pituitary, fat deposition, and body weight in bird migration. *Condor*, 47:95-127.
1952. The occurrence and regulation of the refractory period in the gonadal and fat cycles of the junco. *Jour. Exper. Zool.*, 121:311-325.
1959. Ecologic and physiologic factors in the regulation of spring migration and reproductive cycles in birds. *In Comparative Endocrinology Proc. Columbia Univ., Symp. Comp. Endo.*, edited by A. Gorbman (John Wiley and Sons, New York), pp. 38-70.

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