

EFFECTS OF DURATION AND TIME OF FOOD AVAILABILITY ON PHOTOPERIODIC RESPONSES IN THE MIGRATORY MALE BLACKHEADED BUNTING (*EMBERIZA MELANOCEPHALA*)

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Summary

The effects of the duration and time of food availability on stimulation of the photoperiodic responses (fattening and gain in body mass, and growth and development of testes) were investigated in the migratory blackheaded bunting (*Emberiza melanocephala*). Two experiments were performed. Experiment 1 examined the effects of a reduction in the duration of food supply in buntings that were subjected to long day lengths (16h:8h L:D) and received food *ad libitum* (group I) or for restricted durations, coinciding with the end of the lights-on period, of 8h (group II) and 4h (group III). Buntings of group I gained in body mass, whereas there was a mixed response in group II (half the birds gained and half lost body mass), and all birds of group III lost body mass. There was no effect on testis growth in groups I and II, but testes grew more slowly in group III. Experiment 2 investigated the effects of both the duration and the time of food availability. Of five groups of birds, group I was exposed to an 8h:16h L:D photoperiod, and groups II–V were exposed to 16h:8h L:D. Whereas birds of groups I and II received food *ad libitum*, those of groups III–V were fed only for 5h, at zt 0–5 (group III), zt 5.5–10.5 (group IV) or zt 11–16 (group V), where zt = zeitgeber time and zt 0

refers to the beginning of the lights-on period. Apart from duration, the timing of food availability also had an effect on photoperiodic stimulation under the 16h:8h L:D photoperiod. Birds that were fed *ad libitum* fattened and gained in body mass, whereas among restricted feeding groups, only birds in the group fed during the first 5h (zt 0–5, group III) showed a significant increase in body mass (albeit considerably lower than in the *ad libitum* group). Birds fed during the middle 5h (zt 5.5–10.5, group IV) showed an intermediate response, and those fed during the last 5h (zt 11–16, group V) lost body mass. Testicular growth was suppressed in birds that were fed for 5h in the evening, but not in those fed for the same period in the morning or in the middle of the long day. Taken together, these results show that the duration of food supply and/or the time of day at which food is available affect photoperiodic stimulation of fattening and gain in body mass as well as the growth and development of gonads in the blackheaded bunting.

Key words: body fattening, blackheaded bunting, *Emberiza melanocephala*, food intake, gonadal development, photoperiodic response.

Introduction

In a species that migrates, the regulation of migration and the accompanying metabolic adaptations (e.g. hyperphagia, body fattening etc.) are closely integrated with regulation of the reproductive cycle. For example, the cycles of food intake, body fattening and gonad development are phase-related, and the phase relationship among them is determined by changes in day length (Wingfield and Farner, 1980; Jain and Kumar, 1995; Kumar, 1997). It is believed that day length (photoperiod) has dual actions on the circadian photoperiodic system (Pittendrigh, 1972). It entrains the circadian rhythm of photoperiodic photosensitivity (CRPP) and, if the light period extends into the photosensitive phase (photoinducible phase) of the CRPP, the physiological responses are induced.

In the absence of a direct assay, the rate and magnitude of the photoperiod-induced physiological responses (e.g. fattening

and gain in body mass, and growth and development of gonads in a migratory species) indicate the circadian effects of light on the CRPP. Besides the photoperiod, food cycles (periodic access to food) can also synchronize some circadian functions (for example, circadian activity rhythms in house sparrows, *Passer domesticus*; Hau and Gwinner, 1996), but it is not known if food can exert effects on the CRPP that mediate photoperiodic gonadal responses. To date, the effects of reductions in food supply on body fattening and migratory restlessness during migratory seasons have been studied in a few avian migrants (for details, see Gwinner et al., 1988).

The blackheaded bunting (*Emberiza melanocephala*) is a long-distance palaeartic–Indian migratory small passerine finch (family Emberizidae) that breeds in west Asia and east Europe (approximately 40°N) and overwinters in India

(approximately 25 °N) (Ali and Ripley, 1974). It is seasonal and under natural day lengths (NDL), captive birds show periodic cycles in food intake, body fattening and gain in body mass, testicular growth and development, and plasma levels of luteinizing hormone (LH), testosterone and thyroxine (Jain and Kumar, 1995). The changes in body mass and testicular size occur in parallel, and correspond to the increasing day lengths of spring and early summer, but the cycle of food intake is almost in antiphase (Jain and Kumar, 1995). Also, under artificial day lengths, a long photoperiod ($>11.5 \text{ h day}^{-1}$) stimulates body fattening, gonadal growth and development, and an increase in plasma levels of LH (Tewary and Kumar, 1982; Kumar and Tewary, 1983; Kumar et al., 1993). However, the phase relationships among food intake, body fattening and gonadal growth are not known for artificial day lengths.

In the present study, we investigated whether periodic food availability affects the stimulation of physiological responses in a photoperiodic species. Specifically, we determined if a reduction in the duration and/or a change in the timing of food supply affect body fattening and gain in body mass and the growth and development of testes in photosensitive blackheaded buntings subjected to highly inductive long day lengths.

Materials and methods

Two experiments were performed on the adult male blackheaded buntings (*Emberiza melanocephala* Scopoli) that were caught from the overwintering flocks at 25 °N.

Experiment 1

This experiment examined whether the duration of food availability affects the stimulation of body fattening and gonadal growth and development under long day lengths, and used a batch of birds that were maintained in NDL since 1 February, 1999. The experiment began on 19 April, 1999 (day length=13.8 h, including civil twilight periods). At this time in NDL, birds were slightly fattened (fat score range, 1–3, see below) and of greater mean (\pm S.E.M.) body mass ($35.31 \pm 1.59 \text{ g}$, $N=18$, compared to birds from the same batch maintained on short photoperiods, 8 h:16 h L:D; $26.66 \pm 0.43 \text{ g}$), but the testes were small and unstimulated (testis volume= $0.30 \pm 0.03 \text{ mm}^3$, $N=18$). Three groups ($N=5-6$ per group) were transferred to an 8 h:16 h L:D photoperiod to synchronize them to an artificial photoperiod. After 1 week of 8 h:16 h L:D with an *ad libitum* food supply, all groups were subjected to a highly inductive long day length (16 h:8 h L:D) for a period of 3 weeks but with a differential food supply. Group I received food *ad libitum*; groups II and III received food for a restricted duration. Since buntings do not eat during darkness (V. Kumar, S. Rani and S. Singh, unpublished observations), the duration of food availability for group I birds was taken to be 16 h. For groups II and III, the duration of food availability was reduced to 8 h (zeitgeber time, zt 8–16, where zt 0 indicates the beginning of the lights-on period) and 4 h (zt 12–16), respectively. The timing of restricted food availability

in groups II and III coincided with the end of the lights-on period, and not with the beginning of the lights-on period. This was done to avoid an overlap between the beginning of access to food with the beginning of activity and other light-related behaviours at lights-on (zt 0) in a day-active species like the blackheaded bunting.

Experiment 2

This experiment investigated the effects of the timing of food availability on the stimulation of the photoperiodic responses. The experimental design was similar to that of experiment 1. It began on 19 March, 2000 (day length=13.0 h including civil twilight periods), one month earlier than experiment 1, from a batch of birds that had been maintained in NDL since 10 February, 2000. At this time in NDL, birds were not fattened (body mass= 26.63 ± 0.42 , mean \pm S.E.M., $N=30$) and testes were small and unstimulated (testis volume= $0.30 \pm 0.03 \text{ mm}^3$, $N=30$).

Five groups of birds ($N=6$ in each) were exposed to an 8 h:16 h L:D photoperiod for 1 week, as in experiment 1, and fed *ad libitum*. Group I then remained on this photoperiod, but groups II–V were transferred to 16 h:8 h L:D. Whereas birds in groups I and II were fed *ad libitum*, those in groups III, IV and V were fed only for 5 h (group III at zt 0–5, group IV at zt 5.5–10.5, group V at zt 11–16). The experiment ran for a period of 3 weeks.

During an experiment, birds were held in the photoperiodic chambers illuminated by fluorescent tubes. Food intake was measured daily, although bi-weekly data only are shown in Fig. 1, Fig. 2. Observations on fattening and body mass were recorded weekly, and growth and development of testes were assessed at the beginning and end of the experiments. Food intake was measured as described by Jain and Kumar (Jain and Kumar, 1995). For technical reasons, we measured the food intake for a group of birds ($N=5-6$) that were housed within one wire-mesh cage (45 cm \times 25 cm \times 25 cm) lined with opaque white polythene sheets up to 7 cm above the perch level and placed in steel trays, to facilitate spillage collection. A weighed quantity of food was dispensed into a food cup and, after the period of food availability, food was removed and weighed. In groups with an *ad libitum* food supply, food was dispensed at the time of lights-on, and unconsumed food and faeces were collected just before the next lights-on. This gave the amount of food eaten and the faeces produced by a group in 24 h. In groups with a restricted food supply, food was dispensed only for the specified period during lights-on (i.e. for 8 h and 4 h in groups II and III, respectively, of experiment 1; for 5 h in groups III–V of experiment 2). In these restricted feeding groups, faeces were collected 2 h after the end of the feeding period. We chose the 2 h post-feeding period arbitrarily, although there may be a greater delay between food consumption and faeces production, because during the intervening post-feeding 19 h of different light regimes used for the various restricted feeding groups, light-related behaviours may affect digestive physiology. The difference between the food supplied and that recovered gave the amount

of food consumed (food intake) by a group of birds, and from this the average individual food intake ($\text{g bird}^{-1} \text{day}^{-1}$) was calculated (food intake $\text{g group}^{-1} \text{day}^{-1}$ divided by the number of birds in the group). Faeces were dried and weighed and faeces production as average $\text{g bird}^{-1} \text{day}^{-1}$ was calculated. Utilization efficiency was calculated from these average values for a single bird using the following formula: $[(\text{food intake} - \text{faeces produced}) / \text{food intake}] \times 100$.

Fat deposition was scored on a scale of 0–5 (0=no visible fat deposit, 5=maximum fat deposit all over body; Kumar et al., 1991), and body mass was recorded using a top-pan balance to an accuracy of 0.1 g. The testicular response was assessed by laparotomy under local anaesthesia (Rani, 1999). Briefly, 2% xylocaine (Astra-IDL Ltd. Bangalore, India) was injected subcutaneously, a small incision was made between last two ribs on the left flank and the left testis was located within the abdominal cavity with the help of a spatula. The dimensions of the left testis were recorded and testis volume was calculated from $4/3\pi ab^2$, where a and b denote half of the long and short axes, respectively.

Data are presented as means \pm S.E.M. Where appropriate data were subjected to one- or two-way analysis of variance (ANOVA) with or without repeated measures and followed by *post-hoc* tests. Mean values for the same group before and after a treatment were compared using a paired *t*-test. On some occasions, the means of two groups were also compared using Student's *t*-tests. Significance was taken at $P < 0.05$.

Results

Experiment 1

Food intake appeared to be related to the duration of food availability. Buntings fed *ad libitum* had the highest food intake (Fig. 1A); utilization efficiency did not differ markedly among the three groups, ranging from 86% to 92%. There was a significant ($P < 0.05$, paired *t*-test) gain in mass of birds that were fed *ad libitum* for 21 days, but not for birds that were fed for 8 h or 4 h (Fig. 1B). The mean change in body mass of birds in the *ad libitum* group was also significantly greater ($P = 0.0049$, unpaired *t*-test) than that in the group fed for 4 h, in which most (four out of five) birds lost body mass ($P = 0.0711$; day 0 *versus* day 21, paired *t*-test). There was a mixed response in the 8 h group (half the birds gained and half lost body mass).

Testes were enlarged in all groups (Fig. 1C), although full gonadal development occurred only in groups that were fed *ad libitum* or for 8 h. In the 4 h group, most (four out of five) birds showed a small to moderate response; mean testis volume of this group was significantly lower than that of the other two groups (one-way ANOVA: $F_{2,14} = 8.702$, $P = 0.0035$; $P < 0.05$, Student–Newman–Keuls test).

Experiment 2

There was a clear difference in food intake between short- and long-day control birds. Food intake in long-day birds was higher, even though both groups had an *ad libitum* food supply.

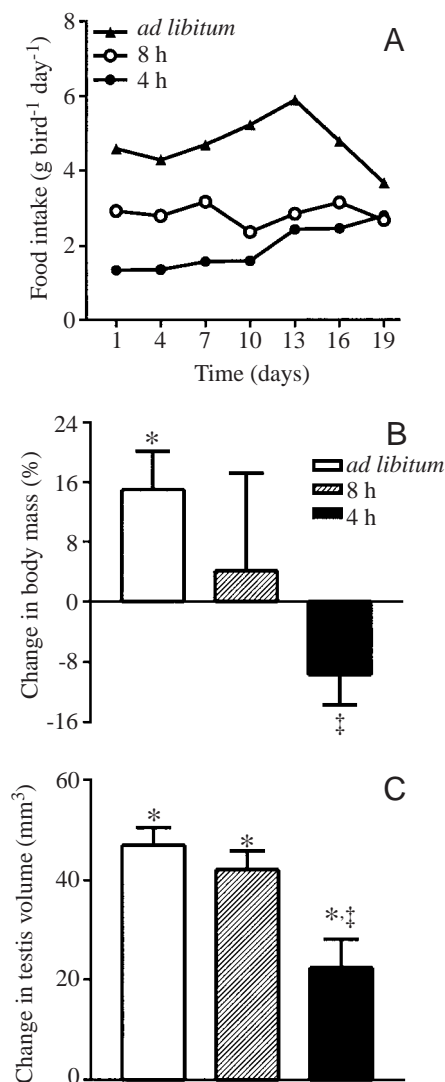
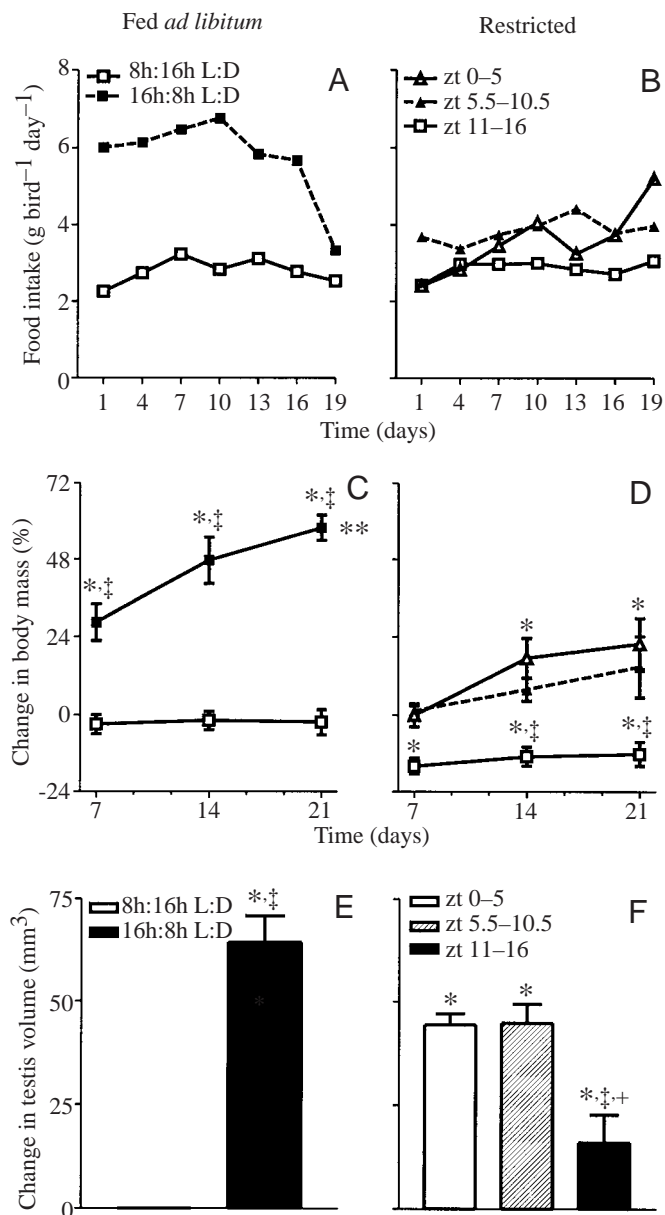


Fig. 1. Results from experiment 1. (A) Food intake, (B) body mass on day 21 (shown as per cent change relative to values on day 0), and (C) change in testicular volume on day 21 relative to that on day 0. At the beginning of the experiment, testes of birds in all groups were minimum in size (approximately 0.33 mm^3). Photosensitive birds ($N = 5-6$ per group) were exposed to a 16h:8h L:D photoperiod for 3 weeks and received food *ad libitum* or for 8 h or 4 h only. Values are means \pm S.E.M. except in A where food intake $\text{bird}^{-1} \text{day}^{-1}$ refers to value calculated from a group value, obtained from a group of birds ($N = 5-6$) housed within one cage (see Materials and methods). * indicates a significant difference ($P < 0.05$) from day 0 values (paired *t*-test); † indicates a significant difference compared to the *ad libitum* group at day 21.

No such difference was found among the three groups that were fed for a restricted period of 5 h at different times (Fig. 2A,B). The utilization efficiency of different groups did not differ: that of the two groups (8h:16h and 16h:8h L:D) on the *ad libitum* food supply ranged between 84% and 88%, and was very similar for at least the first 10 days of the experiment. Similarly, the utilization efficiency of the three groups fed for 5 h ranged between 82% and 88% during the experiment.



The body fattening response was as expected. Birds under long, but not short, day lengths fattened (3.75 ± 0.36 , $N=6$) and gained body mass (Fig. 2C). Among the restricted feeding groups, however, there was less or no fattening (fat score range 0–3) and gain in body mass (Fig. 2D). Birds that were fed during zt 0–5 fattened (fat score, 1.25 ± 0.75 ; $N=6$) and gained body mass ($F_{3,9}=8.441$, $P=0.0055$, one-way repeated-measures ANOVA), and birds that were fed during zt 11–16 lost body mass ($F_{3,12}=15.50$, $P=0.0002$, one-way repeated-measures ANOVA). Birds that were fed during zt 5.5–10.5 showed an intermediate response: slight fattening (fat score, 0.75 ± 0.31 ; $N=6$) but no significant mean gain in mass ($F_{3,15}=2.401$, $P=0.1084$, one-way repeated-measures ANOVA). Two-way ANOVA also indicated a significant difference at all levels (food availability, experiment duration, and the interaction term) in the mean body mass among the four groups (groups

Fig. 2. Results from experiment 2. (A,B) Food intake, (C,D) body mass (shown as per cent change relative to values on day 0), and (E, F) change in testis volume after 21 days relative to that on day 0. At the beginning of the experiment, testes of birds in all groups were minimum in size (approximately 0.33 mm^3). Of five groups of photosensitive birds ($N=6$ in each), group I was exposed to an 8h:16h L:D photoperiod and fed *ad libitum*, and groups II–V were subjected to 16h:8h L:D photoperiod and given an *ad libitum* or restricted (5 h) food supply: group II, *ad libitum*; group III, zt 0–5; group IV, zt 5.5–10.5; group V, zt 11–16; zt=0 indicates the beginning of the lights-on period. The experiment ran for a period of 3 weeks. Values are means \pm S.E.M. except in A and B, where food intake $\text{bird}^{-1} \text{ day}^{-1}$ refers to values calculated from a group value, obtained from a group of birds ($N=5-6$) housed within one cage (see Materials and methods). * indicates a significant difference ($P<0.05$) relative to the value on day 0 (one-way repeated-measures ANOVA or paired *t*-test); ** indicates a significant difference within a group as a function of the duration of the experiment (one-way repeated-measures ANOVA); † indicates a significant difference between groups on the indicated days; + indicates a significant difference with respect to the long-day controls.

II–V) that received an identical photoperiod (16h:8h L:D) but a differential food supply (food availability: $F_{3,68}=30.68$, $P<0.0001$; experiment duration: $F_{3,68}=9.142$, $P<0.0001$; food availability \times experiment duration: $F_{9,68}=4.439$, $P<0.0001$).

A similar response pattern was found for the photoperiodic induction of testes development (Fig. 2E,F). Testes recrudesced in all birds, but the magnitude of response was different in different groups. Testes were fully grown in long-day (control) birds with an *ad libitum* food supply (Fig. 2E). Among the restricted-feeding groups, birds fed for 5 h in the morning (zt 0–5; group III) and in the middle (zt 5.5–10.5; group IV) of the long day had also fully grown testes, albeit slightly smaller in size than in long-day controls ($P>0.05$, Student–Newman–Keuls test; Fig. 2E,F). In birds that were fed for 5 h in the evening (zt 11–16; group V), testes were not fully grown and were significantly smaller in size than in the other photostimulated groups ($F_{3,16}=12.04$, $P<0.05$, one-way ANOVA and Student–Newman–Keuls test).

Discussion

The present study clearly implicates a role of food in the photoperiodic induction of physiological responses in a long-day species. Despite being exposed to a highly inductive day length (16h:8h L:D), buntings did not fatten, but rather lost body mass if they were fed for a restricted period (Fig. 1B, Fig. 2D). There was no effect, however, on utilization efficiency, suggesting that the biological machinery involved in the metabolism of food was not affected by food deprivation.

It appears that the restricted feeding regimes could not meet the costs of physiological events like lipogenesis and/ or gametogenesis induced by long day lengths. It is known that a reduction in the food supply reduces the degree of storage of extra calories as fat and subsequently causes decrease in body

mass in migratory species (e.g. spotted flycatcher *Muscicapa striata*; Biebach, 1985; garden warbler *Sylvia borin*, Gwinner et al., 1985). In experiment 1 of the present study, an 8 h food-availability period coinciding with the end of the lights-on period had an intermediate effect on fattening and body mass gain, but the growth and development of the testes was unaffected (Fig. 1B,C). However, a 4 h food-availability period at the same phase clearly attenuated the rate and/or magnitude of gain in body mass and testicular growth (Fig. 1B,C). These observations suggest that an 8 h food supply coinciding with the end of lights-on, under long day lengths, is close to the critical period of food supply that can sustain gonadal growth and development. Also, the physiological processes involved in fattening and body mass gain are directly influenced by a reduced food availability, and the effect on gonadal growth is presumably a consequence of the reduced extra energy resources available.

Of more interest is the observation arising from experiment 2 (Fig. 2) that the timing of restricted food availability influences the rate and/or magnitude of photoperiodic induction. Food given for 5 h at different times during lights-on produced differential long-day effects (Fig. 2D,F). Whereas no ($P > 0.05$) attenuation was noticed when food was given in the morning (zt 0–5) or the middle of the long day (zt 5.5–10.5), photoperiodic induction was greatly attenuated when food was given in the evening at zt 11–16 (Fig. 2E,F). We do not know precisely how a reduction in food supply exerts a time-dependent effect. However, one explanation could be that birds with different duration of pre-feeding food deprivation during the lights-on period will have different amounts of energy available to meet the physiological costs associated with performing light-related behaviours. If pre-feeding food deprivation in the lights-on period, for example, induces higher locomotor activity (which is costly) compared to the same duration of post-feeding food deprivation, then the total energy expenditure over 24 h will be higher in the group experiencing longer pre-feeding starvation. As a result, less energy will be available to meet physiological costs such as the reproductive cost in birds with a long pre-feeding food-deprivation period; the longer the period of pre-feeding starvation, the greater the magnitude of the effect. In experiment 2 of the present study, a 5 h supply of food given at three times of the long day resulted in three different periods of pre-feeding and post-feeding food deprivation in relation to the beginning of lights-on and these had different photoperiodic effects (Fig. 2D,F). Buntings of group V with the longest pre-feeding (11 h) and no post-feeding lights-on food-deprivation period lost fat and body mass and showed attenuated testicular growth (Fig. 2D,F). By contrast, birds of group III with no pre-feeding and the longest post-feeding (11 h) lights-on food-deprivation period had significant increases in body mass and full testicular growth. Group IV with a 5.5 h pre-feeding and post-feeding lights-on food-deprivation period showed little reduction in fat storage and no effect on testicular growth. In the absence of measurements of any other light-related behaviour, e.g. locomotor activity, in the

present experiments, however, the above explanation remains purely speculative.

Another explanation arises if food availability at different times of the day induces differential effects on the endogenous clock. It is known that periodic food availability can synchronize circadian functions (Hau and Gwinner, 1996), which are photoperiodically regulated. There may be close integration of light and food cues in the timing of a physiological event, such as the growth and development of gonads. This has been shown in the regulation of the annual testicular cycle of an opportunistic breeder, the red crossbill *Loxia curvirostra* (Hahn, 1995). A close integration of light and food cues at the physiological level might mean that there exists an endogenous circadian rhythm of feeding (CRFE), similar to that of photosensitivity (CRPP), in the blackheaded bunting also. In the synchronized state, the onsets of the CRFE and CRPP coincide and, hence, feeding occurs at the time when it is most needed to support the physiological events. In *ad libitum* food supply conditions, the synchrony between light and food cues is never lost. But, in situations such as experiment 2 (groups IV and V) the synchrony between the light and food cues is lost. As a result, the initiation of physiological responses is affected.

A careful comparison reveals a difference in the response to the 16h:8h L:D photoperiod between experiments 1 and 2. Birds of experiment 1 showed a smaller increase in body mass (cf. Fig. 1B, Fig. 2C) and smaller testes (cf. Fig. 1C, Fig. 2E), even though both groups had an *ad libitum* food supply. A relatively smaller gain in body mass in experiment 1 may simply be because of the larger initial body mass of these birds. In late April, when experiment 1 began, most birds had already gained 6–8 g (approximately half of their total expected mass gain under long day lengths) in body mass in response to the increasing day lengths (from 13.0 h to 13.8 h including twilight periods) of March–April. Therefore, upon exposure to a longer photoperiod (16h:8h L:D), these buntings added another 6–8 g and attained maximum fat and body mass. The lower testicular growth observed in experiment 1 could be due to one or both of the following reasons. First, photostimulation of endocrine events, responsible for gonadal growth and initiated by the endogenous clock (e.g. an increase in the circulating levels of gonadotropins), might already have begun at this late date in spring, and exposure to an even longer photoperiod did not augment their secretions. Second, there might be a small change in the responsiveness of the endogenous clock system to light at this time of year. We have discovered very recently that in birds maintained on an 8h:16h L:D photoperiod from February, the testes in birds then exposed to 16h:8h L:D in the middle of April grow more slowly than in those similarly exposed in the middle of March (V. Kumar, M. Misra and S. Malik; unpublished data).

In conclusion, the present findings support the idea that photoperiodism in a seasonally breeding species is affected by food availability. Food can have both direct and temporal (clock) effects. Direct effects are caused because of the unavailability of extra energy during the restricted feeding

regimen to meet the physiological costs. Temporal effects are induced because of asynchrony between light and food cues. A close phase relationship between light and food cues in timing the photoperiod-induced physiological events may clearly have adaptive implications. For example, the presence of food at the beginning of the day (active period), after the no-feeding dark period, is clearly advantageous. In a day-active species like buntings, which are presumably unable to forage during darkness due to photosensory limitations, it is advantageous if feeding coincides with the onset of the day. It would be disadvantageous to have independent regulation of light- and food-controlled physiological processes in these circumstances.

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References

- Ali, S. and Ripley, S. D.** (1974). *Handbook of Birds of India and Pakistan*, vol. 10. Bombay, London, New York: Oxford University Press.
- Biebach, H.** (1985). Sahara stopover in migratory flycatchers: fat and food affect the time program. *Experientia* **41**, 695–697.
- Gwinner, E., Biebach, H. and von Kries, I.** (1985). Food availability affects the migratory restlessness in caged garden warblers (*Sylvia borin*). *Naturwissenschaften* **72**, 51–53.
- Gwinner, E., Dittami, J. P. and Beldhuis, J. A.** (1988). The seasonal development of photoperiodic responsiveness in an equatorial migrant, the garden warbler *Sylvia borin*. *J. Comp. Physiol. A* **162**, 389–396.
- Hahn, T. P.** (1995). Integration of photoperiodic and food cues to time changes in reproductive physiology by an opportunistic breeder, the red crossbill, *Loxia curvirostra* (Aves: Carduelinae). *J. Exp. Zool.* **272**, 213–226.
- Hau, M. and Gwinner, E.** (1996). Food as a circadian zeitgeber for house sparrows: the effect of food access times. *J. Biol. Rhythms* **11**, 196–207.
- Jain, N. and Kumar, V.** (1995). Changes in food intake, body weight, gonads and plasma concentrations of thyroxine, luteinizing hormone and testosterone in captive buntings exposed to natural daylengths at 29°N. *J. Biosci.* **20**, 417–426.
- Kumar, V.** (1997). Photoperiodism in higher vertebrates – an adaptive strategy in temporal environment. *Indian J. Exp. Biol.* **35**, 427–437.
- Kumar, V. and Tewary, P. D.** (1983). Response to experimental photoperiods by a migratory bunting, *Emberiza melanocephala*. *Ibis* **125**, 305–312.
- Kumar, V., Kumar, B. S., Singh, B. P. and Sarkar, A.** (1991). A common functional basis for the photoperiodic mechanism regulating reproductive and metabolic responses in the migratory redheaded bunting. *Period. Biol.* **93**, 169–174.
- Kumar, V. Jain, N., Singh, B. P. and Kumar, B. S.** (1993). Plasma levels of luteinizing hormone in intact and castrated blackheaded bunting (*Emberiza melanocephala*) exposed to stimulatory and nonstimulatory photoperiods. *Reprod. Nutr. Dev.* **33**, 143–150.
- Pittendrigh, C. S.** (1972). Circadian surfaces and the diversity of possible roles of circadian organization in photoperiodic induction. *Proc. Natl. Acad. Sci. USA* **69**, 2734–2737.
- Rani, S.** (1999). Light relations of the photoperiodic clock in buntings (*Emberiza* sp.). PhD thesis. University of Lucknow, Lucknow, India.
- Tewary, P. D. and Kumar, V.** (1982). Photoperiodic responses of a subtropical migratory finch, the blackheaded bunting (*Emberiza melanocephala*). *Condor* **84**, 168–171.
- Wingfield, J. C. and Farner, D. S.** (1980). Temporal aspects of the secretion of luteinizing hormone and androgen in the white-crowned sparrow, *Zonotrichia leucophrys*. In *Acta 17th Congr. Int. Ornithol.* (ed. R. Nohring), pp. 463–467. Deutsche Ornithologen-Gesellschaft, Berlin.