Environment Conservation Journal 12(3) 23-28, 2011
(ISSN 0972-3099) Abstracted and Indexed

Responsiveness to photostimulation in two passeriform birds

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Received:14.04.2011 **Accepted: 13.07.2011**

Abstract

Two studies were performed to analyze the photoperiodic interaction of testicular growth in brahminy myna and weaver bird. In the first study, birds were exposed to stimulatory long day lengths (15L:9D) and natural day length (NDL) for 60 days. The second study investigated the interpretation of a light pulse as 'morning (entraining)' or 'evening (inducing)' depends on the time during night at which they fall. Five groups (6L:6D:1L:11D, 6L:13D:1L:4D, 11L:13D, 13L:11D and NDL respectively) of birds were exposed under skeleton and complete photoperiods for two months. Body mass and testicular volume was measured on monthly intervals. In the first study, testicular volume among both groups (15L:9D) and (NDL) gradually increased, but more inductive effect was found in 15L:9D. In the second study more induction occurred in testicular volume of groups 6L: 6D: 1L: 11D and 13L: 11D with different magnitude as if it was exposed to long days. Taken together, results demonstrate that birds were sensitive to the stimulatory photoperiod and strongly show that brahminy myna and weaver bird at 29^{0}N, 77^0 45'E latituderesponded similar to the populations living at higher latitudes and these species use the photoperiodic cues from the environment to regulate their reproductive cycles.

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Keywords:*Body mass, testes, circadian rhythm, complete and skeleton photoperiod*

Introduction

Day length regulates seasonal responses in many vertebrates, including several songbird species. The cycle of growth–regression–refractoriness can be reproduced under laboratory conditions by exposing birds to long days. Few studies suggest the role of light intensity and photoperiod in initiation of gonadal growth and development (Rani *et al*. 2009; Rani *et al*. 2007; Budki*et al*., 2009; Dixit and Singh, 2011). Long day lengths (15L:9D) induce gonadal growth and development, while short day lengths (photoperiods less than 9h per day) are ineffective or inhibitory in many series of investigation in Indian species; Indian weaver bird (Singh and Chandola, 1981; Rani, *et al*., 2007; 2009), redheaded bunting (Tewary and Tripathi, 1983; Budki*et al*., 2009), blackheaded bunting (Tewary and Kumar, 1982; Misra*et al*., 2004). In this study, we investigated the comparison between NDL (~14L to ~15L) and LDL (15L:9D) in two subtropical Indian species on reproductive cycle. So it was interesting that when we compare the two resident Indian species in breeding season in NDL and LDL (artificial photoperiod) and both species breeding cycle are relatively close and faces the same environmental condition for survival. And also we have aimed to address this question that do different photoperiodic species have 'different reaction norms' to stimulatory day lengths?

Skeleton photoperiods or Scotophase experiments shows the effects of a light pulse which would vary depending where it falls in a 24 h day. This is best illustrated by the ability of two short light pulses introduced at a fixed hour in the circadian rhythm of photoperiodic photosensitivity (CRPP) to induce the metabolic and gonadal functions (Kumar and Follett, 1993; Tewary and Kumar, 1983). It is suggested that in a two pulse light:dark (LD) cycle paradigm (two light pulse at fixed intervals in a 24hr LD cycle), that constitutes a 'skeleton' photoperiod (SKP), the first (usually

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longer) light pulse (main photoperiod) entrain CRPP and the second (usually shorter) light pulse falling in the night (Φi) induces photoperiodic responses (Bhardwaj,*et al*., 2006). Thus, two light periods tend to simulate the effects of corresponding long light period. In our study, the first light period (called the entraining light pulse, E-pulse) given early in the subjective day 'entrains' the CRPP, and photoinducible phase (Φi) begins some 'fixed' hours later in day. Thus, the E-pulse entrains photoperiodic response curve and decides the timing of the Φi. Light coinciding with the Φi initiates photoperiodic reaction. Later, these experiments were applied on a number of species including *Passer domesticus* (Kumar *et al*., 2004, Anushi and Bhardwaj, 2006, 2010), *Coturnixcoturnix japonica* (Follett and Sharp, 1969; Wada, 1981), *Passer montanus* (Lofts and Lam, 1973), *Ploceusphilippinus* (Singh and Chandola, 1981), *Carpodacuserythrinus* (Kumar and Tewary, 1982), *Emberizamelanocephala* (Tewary and Kumar, 1984) and *Sturnuspagodarum* (Kumar and Kumar, 1993). Singh *et al.* (2002), also found the effects of duration and the timing of the E-pulse on photoperiodic induction in the blackheaded bunting.So, in this investigation we answered that 'Is the interpretation of a light pulse as 'morning (entraining)' or 'evening (inducing)' depends on the time during night at which they fall'. In other words, does position of Φi determine the effects of light pulse? We introduced 1 h light pulse at the beginning of night or after midnight and measure the photoinduction to determine the entrainment/re-entrainment pattern in weaver birds.

Materials and Method

Birds were procured from local animal catchers (Meerut 29^0N) in the month of early May 2008 and were acclimatized to captive conditions under natural day lengths (NDL) for a period of two weeks after they were exposed to experimental conditions. First study was performed on adult male brahminy myna (*Sturnuspagodarum*) and baya weaver (*Ploceusphilippinus*) which were divided into four groups (n=6) and were exposed to 15L:9D and NDL.

The second study was performed in February 2008 on adult male brahminy myna which were grouped into five (n=6) and were exposed to 6L:6D:1L:11D (group 1), 6L:13D:1L:4D (group 2), 11L:13D (group 3), 13L:11D (group 4) and NDL (group 5). Day and night situation was provided by artificial illumination of CFL lamps (Phillips) providing cold white light at an intensity of ~500 lux at perch level by switching 'on' and switching 'off' by automatic time switches (Muller clock). Food and water were provided *ad libitum* condition to all groups in both studies.

Body mass was recorded using top pan balance on an accuracy of 0.1g.The dimensions of the left testis were recorded, and testis volume was calculated from formula $4/3лab^2$ where a and b denote the half of the long (length) and short (width) axis, respectively. The data from the experiments was analyzed by one-way repeated measure analysis of variance (one-way RM ANOVA). Newman-Keuls Multiple Comparison Tests compared different means if ANOVA indicated the significance of difference between mean values. Significance was taken at $P < 0.05$.

Results and Discussion

Results of first study are shown in figure 1. There was no significant change in body mass of brahminy myna exposed to 15L:9D, but significant change in weaver birds exposed to 15L:9D and NDL. (fig. 1a and c) [$F_{2,8}$=0.6314, P=0.6540, (15L:9D) brahminy myna, $F_{2,8}$=4.470, P=0.0344, (15L:9D) baya weaver and $F_{2,8}$ =0.6914, P=0.0344, (NDL) baya weaver]. There was significant change in NDL birds of brahminy myna [$F_{2,8}$=17.27, P=0.0005] (fig.1 a).Mean testicular volume in the group exposed to 15L:9D gradually increased throughout the experiment in both species (fig. 1b and d). There was significant increase in testis volume of brahminy myna and baya weaver, subjected to 15L:9D and NDL [$F_{2,8}$=142.6, P<0.0001, (15L:9D) brahminy myna $F_{2,8}$=16.36, P=0.0006, (NDL) brahminy myna; $F_{2,8}$=40.25, P<0.0001, (15L:9D) baya weaver and $F_{2,8}$=18.14, P=0.0004, (NDL) baya weaver; One-way RM ANOVA]. Results of second study are shown in figure 2. Mean body mass among all groups gradually increase throughout the study with different magnitude (fig. 2a and c). The mean body mass among the five groups had significantly different [group 1: $F_{2,8}$=21.24, P=0.0006 (6L:6D:1L: 11D); group 2: $F_{2,6}$=14.39, P=0.0051 (6L:13D:1L:4D); group 3: $F_{2,6}$=4.21, P=0.0053

(11L:13D); group 4: $F_{2,6}$=20.06, P=0.0022 (13L:11D); group 5: $F_{2,6}$=40.01, P=0.0003 (NDL), One-way RM ANOVA]. Figure (2b) shows that mean testicular volume in group 1 and 4 gradually increased by day 30 and then significantly increased on 60 days [$F_{2,8}$=671.2, P<0.0001, (6L:6D:1L:1D) group 1 and $F_{2,6}$ =46.62, P=0.0002, (13L:11D) group 4]. In group 2 (6L:13D:1L:4D), marginal response occurred on day 30 and in group 3 (11L:13D) on day 60, [fig. 2d ($F_{2,6}$=3.479, P= 0.0993, (6L:13D:1L:4D) group 2 and $F_{2,6}$ = 5.537, P = 0.0434, (11L:13D) group 3; One-way RM ANOVA]. There was no photoinduction occurred in the group exposed to NDL throughout the experiment in comparison to group 1 and group 4. More induction occurred in the groups exposed to 6L:6D:1L:11D and 13L:11D with different magnitude.

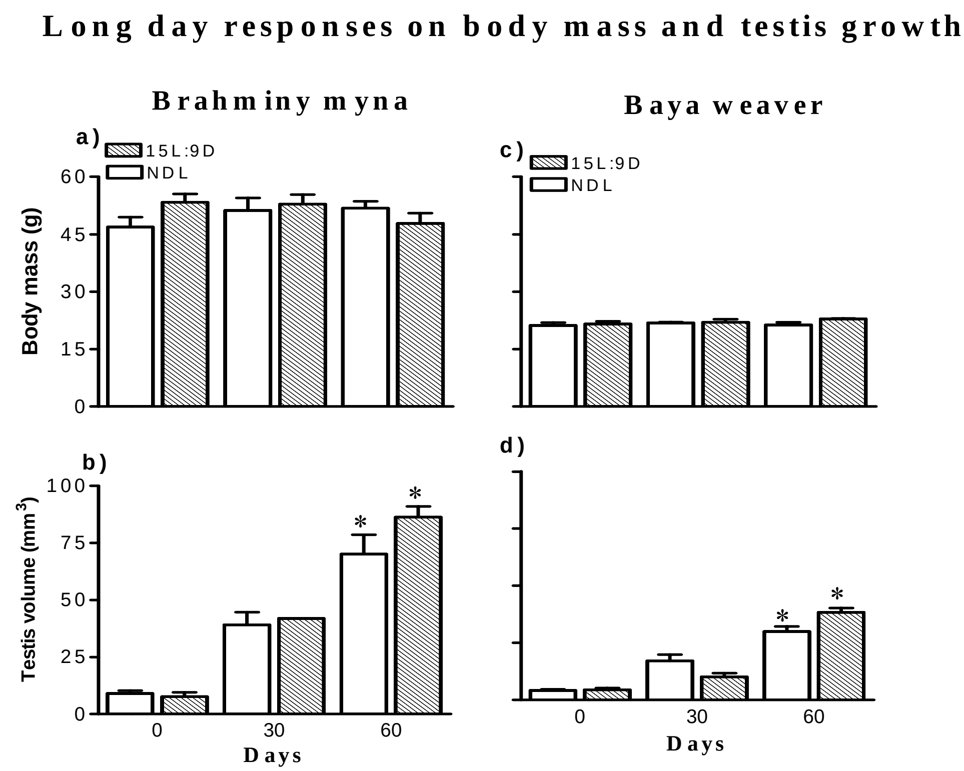

Fig. 1: Results of body mass and testicular volume of brahminy myna (fig. a and b) and baya weaver (fig. c and d) subjected to long day lengths (15L:9D) and natural day lengths (NDL). Asterisk indicates the significance of difference at P<0.05.

In the first study we investigated that the photo-responsiveness of myna and baya weaver under long day lengths to test the photoresponsiveness of neuroendocrine system that regulates reproductive response under long day lengths. Myna and baya are long day breeders and observe the changes in the photoperiod occurring in the nature. There was gain in body mass up to 60 days and testis attaining a peak value in both brahminy myna and baya weaver subjected 15L:9D and NDL groups. The annual cycles of gain in body mass and testes in brahminy myna correspond to increasing day lengths of spring and summer, similar to a number of temperate and tropical/subtropical species (Kumar and Tewary,1983; Dittami and Gwinner, 1985; Kumar and Kumar, 1991, 1993; Deviche and Small, 2001). Male brahminy myna exhibited a seasonal change in its responsiveness to long day lengths, which is comparable to that reported in the Indian weaver bird (Singh and Chandola, 1981). This suggests that under long day lengths induction of a photoperiodic response was faster. A similar photoperiodic response to such photoperiods (9L, 12L and 15L) is reported in another species, the Indian weaver bird, at 25^0N, 83^0E that often shares habitat with the house sparrow. A long day species usually do not show gonadal response under short photoperiods (light below critical day length) and this indicates the importance of photoperiodic cues over an endogenous circannual rhythm in control of reproductive cycle of tree sparrows. On the other hand, gonadal recrudescence under short day length in a long day breeder may be the consequence of seasonal rhythm rather than of the photoperiod. The present results of study 1 shows that both species under 15L:9D and NDL remained stimulated during the entire duration of experiment and thus it indicates that they were sensitive to the stimulatory effects of these photoperiods. Male brahminy myna and baya weaver exhibits similar reaction norms to long days. The results from both the species also suggest that there is no difference in the responsiveness of two photoperiodic species and survive better in natural environment.

Skeleton and Complete photoperiodic effect in brahminy myna

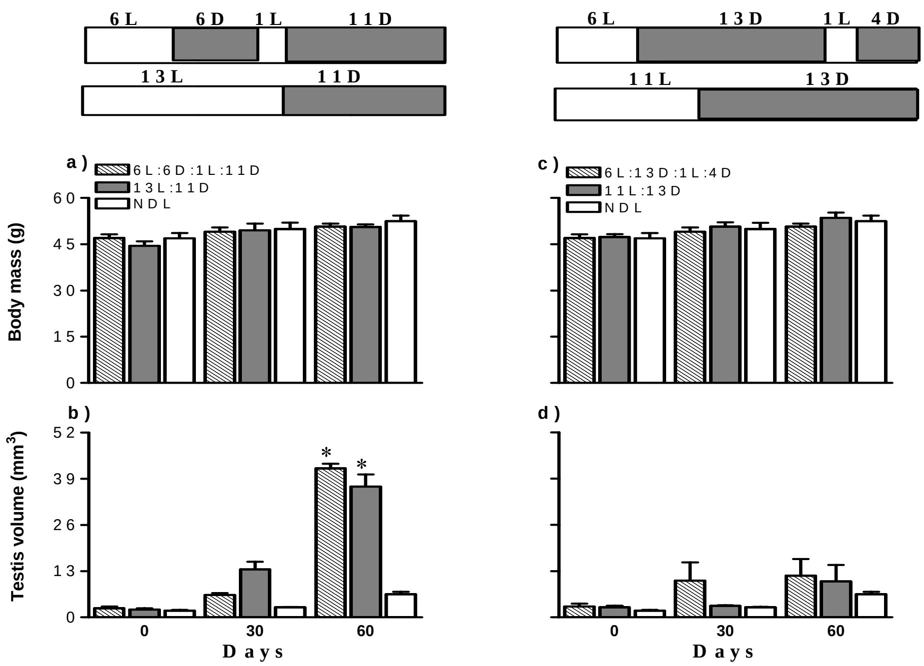

Fig. 2: Results of body mass and testicular responses in brahminy myna subjected to skeleton and complete photoperiods 6L:6D:1L:11D and 13L:11D (fig. a and b) and 6L:13D:1L:4D and 11L:13D (fig. c and d). Asterisk indicates the significance of difference at P<0.05.

In second study a long day response is believed to result from the interaction of long light pulse (LLP) simultaneously with two different phases of the circadian rhythm of photoperiodic photosensitivity (CRPP). This is best illustrated by the ability of two short light pulses introduced at fixed hour in the CRPP to induce the metabolic and gonadal functions (Follett, 1984; Kumar and Follett, 1993; Tewary and Kumar, 1983, 1984; Kumar, 1986, 1988). It is suggested that in a two-pulse light: dark (LD) cycle paradigm (two light pulse at fixed intervals) in a 24 h (LD) cycle, that constitutes a 'skeleton' photoperiod (SKP), the first (usually longer) light pulse (main photoperiod) entrain CRPP and the second (usually shorter) light pulse falling in the night induces photoperiodic responses. Considered together, this means that a SKP will be as effective as a single complete photoperiod (CP, a single continuous light period in a 24 h LD cycle) in influencing the photo-neuroendocrine system in a photoperiodic species.The traditional view of how skeleton photoperiods can stimulate long days is based upon concept of how circadian rhythms might be used as a day length-measuring device. Normally, this coincidence occurs only at one season of the year when the day lengths are of sufficient length to engage the photosensitive phase, but certain skeleton photoperiods can mimic this situation, one of the light pulsecoinciding with the period of peak photosensitivity. The present results on the effect of complete photoperiods (CP) and skeleton photoperiods (SKP) in altering the timing of spontaneous regression. The present data supporting that birds were the photosensitive and when exposed to complete and skeleton photoperiods, their exposure to 6L:6D:1L:11D and 13L:11D did evoke a long day response in brahminy myna (fig 2b) the reason of this behind, because the dark pulse was short duration (6D) so the birds read 6L:6D:1L:11D photoperiod as 13L photoperiod. There was no photoinduction occurred in the group exposed to NDL because the study period was non-breeding phase for brahminy myna. More induction occurred in the groups exposed to 6L: 6D: 1L: 11D and 13L: 11D with different magnitude (fig. 2b) and marginal response occurred in the group exposed to 6L: 13D:1L:4D and 11L:13D (fig. 2d) and mean body mass among all groups gradually increased throughout the experiment (fig. 2a and c). In skeleton photoperiod (6L:6D:1L:11D) the dark pulse was short duration (6D) so the birds entrained according to 13L but in skeleton photoperiod (6L: 13D: 1L: 4D) the dark pulse was longer duration (13D) so the birds reads its, short day length. Result of this study shown that the gonadal response occurred in 13L:11D photoperiod. So, 13L:11D is stimulatory and 11L:13D is non-stimulatory in brahminy myna. Considering together, studies suggest that both birds are photosensitive for long day lengths.

Acknowledgement

Generous financial support through research grant (SR/SO/AS/36/2006 dated 18.10.2007)to SKB from the Department of Science and Technology, New Delhi is gratefully acknowledged.

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