Environmental and Experimental Biology (2023) 21: 27–32
http://doi.org/10.22364/eeb.21.04

Original Paper

Study on photoperiodic modulation of the female fertility of *Drosophila biarmipes*

ISSN 2255-9582

Ahmed Malik Shamim[1]*, Zarin Sana Sayed[2]

[1]Department of Zoology, Progressive Education Society's Modern College of Arts, Science and Commerce Ganeshkhind 16, Pune, India
[2]Department of Zoology, University of Kashmir Hazratbal Srinagar 190006, India

*Corresponding author, E-mail: drshamim530@gmail.com

Abstract

Various attributes of light, including the intensity, spectral composition and duration of the photophase, have been documented to have remarkable influence on all aspects of behaviour and physiology of eukaryotic experimental models. This hypothesis was investigated in the present study wherein the effects of photoperiod varying from 6 to 18 h of 24-h light-dark (LD) cycles was examined on the lifetime fertility (i.e., the viable F_1 progeny) of females of the Indian fruit fly, *Drosophila biarmipes*. The flies were subjected to six types of 24-h LD cycles (light at 150 lux and dark at 0 lux in all LD cycles) in which the photoperiod varied from extremely short (6 h) to extremely long (18 h) duration. The female fertility was the highest in photoperiods of 10, 12 and 14 h, moderate in 8 and 16 h and the lowest in 6 and 18 h. These results indicate that the performance of the circadian system is at its optimum when it is maintained in zeitgeber cycles wherein the photoperiod is close to that of the natural day of the tropical or sub-tropical region from where the given species or strain originated. This study has great implication in predicting the insect abundance at a particular season of the year when the photoperiod is conducible for optimum reproduction. Such premonition should help the local administrative bodies to take appropriate control measures against the given insect pest and vector insects.

Key words: circadian, *Drosophila*, fertility, photoperiod, reproduction.
Abbreviations: CT, circadian time; DD, continuous darkness; LD, light-dark; LED, light emitting diodes.

Introduction

Exogenous time-synchronizers or zeitgebers (time givers, German term), like the cycles of light-dark, temperature, magnetic field etc., are known to modulate almost all features of circadian physiology of most eukaryotic experimental models (Pittendrigh 1981). Cycles of light-dark (LD) are regarded as the most efficient zeitgeber in influencing numerous attributes of physiology of animals. Three attributes of light namely, the intensity, spectral composition and duration of photophase or scotophase are known to affect the waveform, amplitude and the phase of the biological rhythms, that in turn can influence several aspects of metabolism, physiology, behaviour, molecular biology etc. of the eukaryotic experimental models like rats, mice, hamsters, fruit flies, etc. (Saunders 2002; Johnson et al. 2003). For example, a protracted photoperiod was unequivocally shown to lengthen duration of the perch-hopping activity of the finches, which affected other attributes of the circadian rhythmicity (Aschoff, Wever 1965). Photoperiod of varying duration changed the fundamental properties of the eclosion rhythmicity of the fruit fly, *Drosophila pseudoobscura* (Pittendrigh 1981).

Similarly, the circadian rhythm of adult locomotor activity of the temperate species of fruit fly, *Drosophila melanogaster* was modulated by varying the duration of the photophase, wherein these results were attributed to alteration in the expression of clock and timeless genes (Shafer et al. 2004; Rieger et al. 2007; Rieger et al. 2012). Varying photoperiod was also reported to change the ratio of the activity phase to rest phase of the cricket, *Gryllum bimaculatus*, however, the period of free-running rhythmicity in continuous darkness was unaltered (Koga et al. 2005). These results were attributed to the speculative existence of two independent mechanisms, which might govern the waveform of the activity rhythm during entrainment and the free-running rhythmicity in continuous darkness (DD). Photoperiod has been documented to affect the circadian entrainment in which the underlying pacemakers play a key role and also the masking effects on the activity rhythm in which the circadian pacemakers are completely bypassed. These claims were substantiated by demonstrating the effects of varying photoperiods on the circadian as well as the masking peaks of the activity rhythm of the onion fly, *Delia antiqua* (Arai, Watari 1997). There are a few interesting studies describing the effects of photoperiods on the pupal eclosion of a few Indian species of *Drosophila*. For example, two attributes of eclosion rhythmicity of the Indian species of the fruit fly, *Drosophila ananassae* were altered by varying photoperiods

(Joshi, Gore1999).

Moreover, the photoperiods of the entraining light-dark cycles exhibited after-effects on the period of free-running rhythm in subsequent continuous darkness. Apart from the eclosion rhythm, which is expressed during the larval or developmental stage, photoperiods profoundly altered the oviposition rhythm that is expressed during the adult stage, in females of the high altitude Himalayan strains of *D. ananassae* (Satralkar et al. 2007). The after-effects of such a photoperiod on the period of free-running rhythm of oviposition were even observed in subsequent constant darkness. The underlying pacemakers controlling the eclosion rhythm of the high altitude Himalayan strains *D. ananassae*, however, are not affected by the photoperiods of entraining light-dark cycles or intensity of photophase, as the environmental temperature had taken the precedence over the ambient photic stimuli as the results of natural selection at the high altitude (Khare et al. 2004). Other than the photoperiod, the dark phase or scotoperiod of the entraining light-dark cycles may change the basic properties of the overt rhythms of *Drosophila*. For example, the dimly lit scotoperiod (9 to 15 h) selectively altered almost all attributes of the entrainment of the locomotor activity rhythm of the males of *Drosophila jambulina* and subsequent free-running rhythmicity of the locomotor activity rhythm of *D. jambulina* (Thakurdas et al. 2011). Recently, it was elegantly demonstrated that the photoperiod had profound influence on the process of re-entrainment of the adult locomotor activity rhythm of *Drosophila biarmipes*, wherein the accelerating effects of the photoperiod had strong reliance on the direction of the shift of light-dark cycles (Sinam et al. 2013). Thus, the photoperiod of the entraining light-dark cycles has been documented to influence numerous attributes of the circadian physiology of various experimental insect models, however, its effects on the fertility of the Indian fruit fly, *D. biarmipes* have not yet studied. The present study evaluated the effects of changing photoperiods on the fertility of *D. biarmipes*. The objectives of the present investigation were as follows: (i) to determine if the sub-tropical species of fruit fly like *D. biarmipes* responds to an extremely short or long photoperiod, since it is never exposed to such an extreme photoperiod; (ii) to investigate the influence of photoperiods varying from 6 to 18 h on the daily and lifetime fertility of females of *D. biarmipes*.

Materials and methods

Fly strain

The experimental model of the present study was the iso-female culture of the ANR-19 strain of the Indian sub-tropical fruit fly, *Drosophila biarmipes* (Malloch 1924). This strain originated from 121 gravid females captured from 16 different house-gardens at Ahmednagar (19°05'N, 74°48'E) Maharashtra-414001. The larvae and adults were maintained on standard *Drosophila* culture medium that was prepared by the method described below.

Culture medium

The culture medium contained the following ingredients: ground sweet corn (source of carbohydrates), organic brown sugar or jaggery (gud: source of sweetness), agar (solidifying agent), propionic acid (source of fermenting aroma of the ripen fruits), methyl para-benzoate (anti-fungal agent that inhibits the fungal infection for about 10 days) and baker's lump yeast as well as dry yeast grains (larval feed). In addition to the dietary yeast added during the preparation of the culture medium, 6 to 8 yeast grains were sprinkled on the top to augment the reproductive performance of the flies. The culture medium (1 L for 25 bottles) was prepared as follows. Eighteen grams of agar was dissolved in 875 mL boiling water. Then 125 g of unsulfured jaggery/molasses was added and the content was boiled till the rolling point. Corn meal of 125 g and 50 g of dry brewer's yeast was mixed thoroughly in 250 mL of water. This mixture was added to the boiling agar-molasses solution and then heated gently for 15 to 30 min on a low flame. It was cooled to ~ 50 °C and then 5 mL of propionic acid was added. Then warm and semi-molten culture medium was poured in the glass vials or culture bottles (layer of about 2 cm) which were closed with cotton balls so that there would be efficient aeration. A few grains of yeast were sprinkled on top of the culture medium and then a folded filter paper was placed on top of the culture medium so that the extra moisture would be absorbed. The folded filter paper also served as a dry surface for third instars larvae to pupate. Culture medium was replenished at an interval of 10 days to avoid any fungal infection and the subsequent loss of the culture.

Experimental chrono-cubicles

All experiments were performed in specially fabricated windowless chrono-cubicles (3.4 × 4.1 × 3.9 m) fitted with opaque overlapping black blinds and double doors to prevent any entrance of the stray light from outside. Constant room temperature at 25 ±0.5 °C and about 60% relative humidity was maintained by using split-air-conditioning machines (2T-Videocon, India) and humidifiers (NS-301, Devis Appliances, Ahmednagar, India). Cultures were maintained in 24 h cycles of 12 h of white light at 150 lux and 12 h of complete darkness (LD 12:12). This was regarded as the standard rearing condition. Broad-spectrum (365 to 710 nm) white light resembling the diffused daylight was obtained by using ultra-violet-free white light emitting diodes (LED) (6VDC- 0.02W, Legtick Electronics Co. Ltd., Taiwan), which were replaced every 2 months to retain their characteristic luminance with respect to the spectral composition and intensity. Use of incandescent bulbs, halogen lamps, fluorescent tube lights or compact fluorescent lamps was carefully avoided since

such light sources show a peak in the higher wavelength region, i.e., 590 to 640 nm. Moreover, such light sources age comparatively faster than the LED light sources used in the present study. Maintenance or servicing of any emergency event during the dark phase of LD cycle was always carried out by using diffuse red light > 640 nm to which the flies had physiological blindness (Thakurdas et al. 2012).

Photoperiodic enclosures

Seven efficiently ventilated and lightproof plywood boxes (1.2 × 1.5 × 1.1 m) were used as photoperiodic enclosures to accommodate culture bottles containing the adults that were maintained in each of the 7 photoperiods. Light intensity in each photoperiod was at 150 lux at the level of the culture bottles. It was determined by using the photometer (United Detector Technology, Model 40X, Santa Monica, USA). Experimental flies were subjected to six photoperiods as follows: 6, 8, 10, 14, 16 and 18 h per 24 h. The photoperiod in each LD cycle was implemented by connecting LED light sources to electronic time switches (Frontier Digital Timer, TM-619-series, 220/250 VAC-50/60Hz/16A, Taiwan), which had precision of ± 0.14 s per 24 h. Adults maintained in a 12 h photoperiod, i.e., in LD 12:12 cycles were regarded as control flies while the flies maintained in photoperiods other than 12 h were designated as experimental flies.

Evaluation of female fertility

Fertility of the females in the present study was regarded as the F_1 viable progeny. Fertility of fruit flies could also be evaluated by the sperm-count method or the egg-count method. These methods have a few short-comings: the eggs or sperms may or may not be functional gametes (n), i.e., the eggs could be infertile or the sperms could be immobile that render them worthless with regard to the fertility objective of the present study (Bouletreau 1971; Fowler 1973; Pitnick et al. 1995). In contrast, the F_1 viable progeny (2n) is the most dependent fertility index, and shows the successful courtship and mating followed by sequential events like the fertilization, embryonic development, metamorphosis and eclosion (Pitnick, Karr 1998). Moreover, it was easy to count the F_1 progeny precisely as compared to the counting of sperm or eggs, which are of microscopic size. Even the gender of F_1 progeny could be determined, which was not done in the present study since it was not the part of objectives.

Four freshly eclosed males and a virgin female were collected from the standard rearing condition and transferred to a vial (30 mL) containing 5 mL culture medium. There were 31 such vials for each 7 photoperiods. Upon appearance of pupae, in about 6 days in each vial, the parents were transferred to a fresh vial. Such regular transfer of parents was continued till the oviposition female in each vial died. Upon death of the female, any surviving males were discarded immediately. The F_1 progeny was collected till the last individual imago was enclosed. During the course of experimentation if any male died in a vial, then it was replaced by a young one that was collected from the standard rearing condition. This strategy ensured the presence of four males in each vial for one female as mating partners throughout its lifespan. In contrast, if a female would die before it reached the mean lifespan that was specific for the given photoperiod, then the vial was discarded immediately. There were always 3 to 5 such vials in each experiment. Therefore, only 21 vials in each experiment in which the females survived for the duration of the mean lifespan were randomly selected to evaluate the lifetime fertility. This strategy ensured consistency in all experiments. Lifetime female fertility was considered as the mean F_1 viable progeny (± SD, n = 21 females) of the 21 females during the complete lifespan. Similarly, the daily fertility of the females was evaluated by dividing the mean lifetime fertility by the mean lifespan. This strategy served the purpose to pinpoint whether the reduction in lifetime fertility was due to variation in the photoperiod or the mean lifespan in a given photoperiod.

Statistical analysis

Two-way analysis of variance (ANOVA) was carried out on the values of the fertility and photoperiod to determine any significant effects of the photoperiod on the fertility.

Results

Influence of photoperiods varying from 6 h to 18 h on the female fertility of the Indian fruit fly, *D. biarmipes* was investigated in the present experiment. The results showed that the photoperiod strongly affected the daily and lifetime fertility of the females (Table 1, Table 2). There was no clear-cut positive or negative correlation of fertility with increasing photoperiod from 6 to 18 h. However, there was positive correlation (+0.79) when the values of photoperiods ranging from 6 to 12 h were taken into consideration. Similarly, there was a negative correlation

Table 1. Effects of photoperiod on mean daily fertility (mean, ± SD, n = 21 females) and the mean lifetime fertility of females (mean, ± SD, n = 21 females) of the Indian sub-tropical fruit fly, *Drosophila biarmipes*. The daily as well as the lifetime fertility was the highest in 12 h as compared to other photoperiods.

Photoperiod (h)	Daily fertility (number of viable eggs laid per day)	Lifetime fertility (number of viable eggs laid lifetime)
6	15.4 ± 1.9	431.8 ± 19.7
8	18.4 ± 3.1	661.6 ± 12.3
10	20.3 ± 2.5	811.5 ± 22.2
12	27.6 ± 3.2	1188.8 ± 21.3
14	21.2 ± 2.1	802.7 ± 10.6
16	20.2 ± 1.8	668.8 ± 26.2
18	19.1 ± 2.3	443.9 ± 10.3

Table 2. Results of Tukey's HSD post hoc test

Treatments pair	Tukey's HSD Q statistic	Tukey's HSD p-value	Tukey's HSD inference
Daily fertility			
A vs B	2.5015	0.2081121	Insignificant
A vs C	6.8444	0.0010053	** $p < 0.01$
B vs C	4.3429	0.0171805	* $p < 0.05$
Lifetime fertility			
A vs B	5.1772	0.0048242	** $p < 0.01$
A vs C	4.8161	0.0084023	** $p < 0.01$
B vs C	0.3611	0.8999947	Insignificant

(–0.82) when the values of photoperiods ranging from 12 to 18 h were taken into account. It is interesting to note that the three photoperiods, i.e., 8, 10 and 12 h (12 ± 2 h) were indisputably conducible for daily fertility and lifetime fertility. Fig. 1 illustrates the effects of varying photoperiods on the lifetime fertility of the females. The lifetime fertility was found to be optimum in the photoperiods of 10, 12 and 14 h, however, it was moderate in the photoperiods of 8 and 16 h and dismal in the extremely short and long photoperiods of 6 and 18, respectively. It was significantly higher in the 12 h photoperiod than any other photoperiod ($p < 0.01$; Table 2).

Discussion

The performance in most of the physiological processes controlled by the innate circadian pacemakers of the eukaryotic experimental models is the best when the exogenous zeitgebers or oscillators drive the innate circadian system with a frequency that matches the periodicity of the solar day, i.e., 24 h (Pittendrigh, Minis 1972). However, it should be noted that the external light-dark cycles, despite having a 24-h periodicity are not adequate to bring out the best physiological performance. Such 24-h LD cycles must have a tolerable ratio of the photoperiod to scotoperiod. This hypothesis was tested in the present experiment on the Indian fruit fly, *D. biarmipes*. Two important physiological attributes namely, the daily fertility and the lifetime fertility of the females were thoroughly investigated. In nature, the developing stages like the eggs, larvae and pupae or the adults of this species are subjected to 24-h natural LD cycles in which the photoperiods range from ~ 11.5 h in mild sub-tropical winter months to 13.5 h in relatively hot summer months (i.e., 12 ± 1.5 h) at the place of origin of this strain, i.e., Ahmednagar (19°05'N, 74°48'E). Therefore, these flies were subjected to ecologically relevant photoperiods at 10, 12 and 14 h in the present study in which they exhibited the highest fertility (Table 1 and Fig. 1). They were characterized by the moderate fertility when maintained in LD cycles with photoperiods of 8 and 16 h. These results should be attributed to the imposed photoperiods that did not deviate too much from 12 h, i.e., they are within the tolerable range of 12 ± 2.0 h (Joshi et al. 2014). The innate

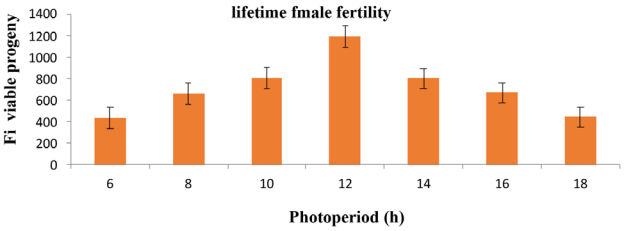

Fig. 1. Effect of the photoperiod varying from 6 to 18 h on lifetime fertility of the females of the Indian fruit fly, *D. biarmipes*.

circadian system that governs these physiological attributes was quite accommodative to embrace such photoperiods close to 12 h (Johnson et al. 2003). However, the fertility of these flies was utterly dismal when they were subjected to an extremely short photoperiod of 6 h, which is similar to the photoperiod of the short days of winter months in the temperate region, or an extremely long photoperiod of 18 h, which resembles the long days of the summer months of the temperate region (Huey 2001). It appears that their inherent temporal programme completely failed to accommodate these extremely short or long photoperiods. These results should be ascribed to the failure of the evolutionary adaptation of this sub-tropical species, which never experiences such extreme photoperiods in the field. This study unequivocally demonstrates that the reproductive performance of the fruit fly, *D. biarmipes* is vulnerable to photoperiodic influences. The fertility could be robust and sustainable in photoperiods close to 12 h, which this species experiences in the wild at Ahmednagar Maharashtra. The moderate degree of fertility in 8 and 10 h photoperiods suggests that the circadian system could be stretched to a certain extent within the limit of the photoperiodic tolerance. The dismal fertility in extreme photoperiods of 6 h or 18 h indicates that such photoperiods could be detrimental to the reproductive performance of this species, as it never encounters such extreme photoperiods in nature (Khare et al. 2007).

The extremely short or long photoperiods are known to act as a weak zeitgeber, resulting in a poor entrainment, damping of the rhythm, arrhythmicity or miserable physiological performance in most attributes (Satralkar et al. 2007; Thakurdas et al. 2011; Sinam et al. 2012). The results of the present study are in agreement with this hypothesis, as extremely short or extremely long photoperiods drastically reduced the fertility of *D. biarmipes*. This might be due to modulation of the underlying pacemakers that control various overt rhythms. In contrast, photoperiods of 10, 12 and 14 h were found to be conducible for fertility, since these photoperiods are very close to the ecologically occurring photoperiods at the origin of this species (Sharma et al. 2012). Other attributes of light like the intensity are also documented to influence several features of the circadian physiology of eukaryotic experimental models (McKenzie 1975). For example, a relatively bright photophase at 300 lux, as compared to the dim one at 30 lux, accelerated the

process of re-entrainment of the adult locomotor activity of *D. biarmipes* following phase delays and phase advances of the implemented light-dark cycles (Sinam et al. 2012). This result was interpreted by pointing out that the adults of this strain of *D. biarmipes* always exist in relatively bright diffused daylight intensity, which changes from ~ 40 lux at the sunrise or sunset to ~ 1900 lux in the forenoon on cloudless days. Apparently, the bright photophase at 300 lux acted as a strong zeitgeber photic signal stimulating a robust bimodality of the locomotor activity, while the dim photophase at 30 lux was interpreted as a weak zeitgeber that could not initiate or sustain the bimodal activity pattern of the locomotor activity rhythm.

The present study on the Indian fruit fly, *D. biarmipes* was basically designed to investigate the effects of the photoperiodic modulation of the circadian pacemakers on female fertility. This strain of *D. biarmipes* is subjected to a natural photoperiods varying from ~ 10.5 h in the mild sub-tropical winter to 13.5 h in the summer months of Ahmednagar. It never experiences photoperiods like 6, 8, 16 and 18 h of the high latitudes or the temperate regions. It would be interesting to find out how the circadian system of this fly copes with such extremely short or long photoperiods, and to determine whether its innate temporal programme governed by the underlying circadian pacemakers is labile or rigid. In this context, this study would be a significant contribution to our existing knowledge on chronobiology. The second aspect of this study would be to see how female fertility is influenced by a changing photoperiods. This has great importance for the present modern human society that is exposed to an extended photoperiod caused by the electric lighting in urban as well as rural regions where the people willingly indulge in a lifestyle beyond the sunset: i.e., they are exposed to light at night which adds to the existing natural photoperiod. This could have devastating consequences on all aspects of human physiology including reproduction, fertility, sperm count, menstrual cycle, abortion, miscarriage, etc. The third relevant contribution of this study could be prediction of insect pest or vector abundance in a particular season of the year. This experiment evaluated female fertility of *Drosophila* at different photoperiods and by implication, can assist the local governing bodies to prepare for insect abundance during a certain season when the photoperiod would be highly conducible for the reproduction of certain insect pest or vector. Apart from above mentioned relevance, this study appears to be the pioneering type with respect to academic enquiry into influence of extremely short or long photoperiods on the fertility of the tropical or a sub-tropical eukaryotic experimental insect model.

Conclusions

The outcome of this study demonstrates that the photoperiod of entraining light-dark cycles profoundly changed the fertility of *D. biarmipes*. This study may have great implication in predicting the photoperiod-dependent fluctuations in populations of beneficial as well as harmful insects in the field at particular times of the year, since the natural photoperiod predictably changes over the seasons.

Acknowledgements

I would like to express my special gratitude and thanks to my supervisor Dr. Dilip S. Joshi, Associate Professor and Head Department of Zoology, Ahmednagar College Ahmednagar -414001 for imparting his knowledge and expertise in this study. I am grateful to Dr. R. J Barnabas, Principal, Ahmednagar College Ahmednagar for providing the necessary infrastructure and laboratory facilities to carry out this research. I also thank the former Vice Principal Dr. S. B. Iyyer and the present Vice Principal Dr. N. R. Somawanshi for their moral support. I am also thankful to Dr. Pooja Thakurdas and S. B. Sinam and Ms. Shweta Sharma for their invaluable help during all stages of the experimentations. I am highly indebted to my parents and other family members for their unwavering moral and financial support without which it would have been difficult for me to complete this work. The experiment was carried out in controlled laboratory conditions to avoid any conflict with ethical standard.

References

Arai T., Watari Y. 1997. Effects of photoperiod and aging on locomotor activity rhythms in the onion fly, *Delia antiqua. J. Insect Physiol.* 43: 567–576.

Aschoff J., Wever R. 1965. Circadian rhythms of finches in light-dark cycles with interposed twilights. *Comp. Biochem. Physiol.* 16: 507–514.

Bouletreau-Merle J. 1988. Biological factors. In: Lints F.A., Soliman M.H. (Eds.) Drosophila *as a Model Organism for Ageing Studies.* Springer, New York, pp. 85–96.

Fowler G. 1973. Some aspects of the reproductive biology of *Drosophila*: sperm transfer, sperm storage, and sperm utilization. *Adv. Genet.* 17: 293–360.

Gibert P., Huey R.B. 2001. Chill-coma temperature in *Drosophila*: effects of developmental temperature, latitude, light intensity, phylogeny. *Physiol. Biochem. Zool.* 74:429–434.

Johnson C., Elliot J., Foster R. 2003. Entrainment of circadian programs. *Chronobiol. Int.* 20: 741–774.

Joshi D., Gore A. 1999. Latitudinal variation in eclosion rhythm among strains of *Drosophila ananassae. Indian J. Exp. Biol.* 37: 718–724.

Khare P., Keny V., Vanlalnghaka C., Satralkar M., Kasture M., Barnbas R., Joshi D. 2004. Effect of temperature, photoperiod and light intensity on the eclosion rhythm of the high altitude Himalayan strain of *Drosophila ananassae. Chronobiol. Int.* 21: 351–363.

Khare P., Keny V., Vanlalnghaka C., Satralkar M., Kasture M., Barnbas R., Joshi D. 2007. Effect of photophase on oviposition rhythm of the strains of *D. ananassae Chronobiol. Int.* 24: 389–405.

Koga M., Hiroshi U., Kenji T. 2005. Photoperiodic modulation of circadian rhythms in the cricket *Gryllus bimaculatus. J. Insect Physiol.* 51: 681–690.

Malloch J. 1924. Two Drosophilidae from Coimbatore. *Mem. Dept. Agric. India Entomol. Ser.* 8: 63–65.

Pittendrigh S. 1981. Circadian surfaces and the diversity of

possible roles of circadian organization in photoperiodic induction. *Proc. Natl. Acad. Sci. USA* 69: 2734–2737.

Pitnick S., Spicer S., Markow T. 1995. How long is a giant sperm? *Nature* 375: 109–109.

Pitnick S., Karr L. 1998. Paternal products and by-products in *Drosophila* development. *Proc. Royal Soc. London B* 265: 821–826.

Pittendrigh S., Minis H. 1972. Circadian systems: longevity as function of circadian resonance in *Drosophila melanogaster. Proc. Natl. Acad. Sci. USA* 69: 1537–1546.

Reiger D., Stanewsky R., Helfrich-Forster C. 2003. Cryptochrome, compound eyes, Hofbauer-Buchner eyelets, and ocelli play different roles in the entrainment and masking pathway of the locomotor activity rhythm in the fruit fly *Drosophila melanogaster. J. Biol. Rhythms* 18: 377–391.

Rieger D., Fraunholz C., Popp J., Bichler D., Dittmann R., Helfrich-Förster C. 2007. The fruit fly *Drosophila melanogaster* favors dim light and times its activity peaks to early dawn and late dusk. *J. Biol. Rhythms* 22: 387–399.

Rieger D., Peschel N., Dusik V., Glotz S., Helfrich-Förster C. 2012. The ability to entrain to long photoperiods differs between 3 *Drosophila melanogaster* wild-type strains and is modified by twilight simulation. *J. Biol. Rhythms* 27: 37–47.

Saunders S. 2002. Circadian rhythms of activity in populations of insects. In: Saunders D.S. (Ed.) *Insect Clocks.* 2nd Ed. Elsevier, Amsterdam, pp. 43–102.

Satralkar M., Khare P., Keny V., Chhakchhuak V., Kasture M., Shivagaje A., Iyyer S., Barnabas R., Joshi D. 2007. Effects of photophase and altitude on oviposition rhythm of the Himalayan strains of *Drosophila ananassae. Chronobiol. Int.* 24: 389–405.

Saunders D. 2002. Circadian rhythms in photoperiodism. In: Steel C., Vafopoulou X., Lewis R. (Eds.) *Insect Clocks.* 2nd Ed. Elsevier, Amsterdam, pp. 339–376.

Shafer O., Levine J., Truman J., Hall J. 2004. Flies by night: Effects of changing day length on *Drosophila's* circadian clock. *Curr. Biol.* 14: 424–432.

Sharma S., Thakurdas P., Sinam B., Joshi D. 2012. Paradoxical masking effects of bright photophase and high temperature in *Drosophila malerkotliana. Chronobiol. Int.* 29: 157–165.

Sharma S., Thakurdas P., Sinam B., Joshi D. 2012. Bright photophase accelerates re-entrainment after experimental jetlag in *Drosophila. Naturwissenschaften* 99: 575–578.

Silber S.J. 1991. Effect of age on male fertility. *Seminars Reprod. Endocrin.* 9: 241–248.

Sinam B., Sharma S., Thakurdas P., Joshi D. 2012. Bright photophase accelerates re-entrainment after experimental jetlag in *Drosophila. Naturwissenschaften* 99: 575–578.

Sinam B., Sharma S., Thakurdas P., Joshi D. 2013. Influence of photoperiod in accelerating the re-entrainment in *Drosophila. Chronobiol. Int.* 29: 1405-1411.

Stover J. 1998. Revising the proximate determinants of fertility framework: what have we learned in the past 20 years? *Stud. Family Plann.* 29: 255–267.

Thakurdas P., Sharma S., Singh B., Vanlalhriatpuia K., Joshi D. 2011. Varying the length of dim nocturnal illumination differentially affects the pacemaker controlling the locomotor activity rhythm of *Drosophila jambulina. Chronobiol. Int.* 28: 390–396.

Received 31 January 2023; received in revised form 18 March 2023; accepted 20 March 2023