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Circadian Rhythm in the Locomotor Activity of a Surface-Dwelling Millipede *Syngalobolus* sp.

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ABSTRACT

Locomotor activity of the surface-dwelling millipede *Syngalobolus* sp. was recorded under laboratory conditions. Infra-red diodes were used to detect the locomotor activity in an oval shaped chamber, which was connected with an event recorder. The results of 11 individuals showed that the millipedes entrained to light/dark (LD12:12 h) conditions with negative phase angle difference (-83.2 ± 24.72 min). The millipedes showed a clear-cut free-running rhythm with a period (τ) of 23.8 ± 1.0 h ($n = 9$) in constant darkness (DD). The period in continuous light (LL) was relatively greater (25.2 ± 0.1 h; $n = 3$) than that in DD.

KEYWORDS: Locomotor activity, free-running, entrainment, millipedes.

INTRODUCTION

Circadian rhythms are ubiquitous in biological systems ranging from unicellular algae to human beings (Aschoff, 1960; Wever, 1979; Brady, 1981; Minors and Waterhouse, 1981; Chandrashekar, 1997; Chandrashekar et al., 1997). These rhythms even persist in constant darkness (DD) and constant light (LL) exhibiting 'free-running' periodicity (Aschoff, 1951). Circadian rhythms in numerous organisms have already been investigated under different light regimes. Among the other organisms, there are a few reports available on circadian rhythms in millipedes. The polydesmoid millipedes have been observed to crawl at twilight hours, just before dawn under field conditions. These millipedes show aggregated or

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clumped distribution of activity and rest time (Bano et al., 1976). Such aggregations may arise due to environmental fluctuation or to social attraction or both factors working together (Southwood, 1966). Bano and Krishnamoorthy (1979) also reported persistence of a bimodal crawling activity of *Jonespeltis splendidus* under laboratory conditions. Under light/dark (LD 12:12 h) conditions, peak activities were observed to fall during dusk hours and before dawn; whereas in constant darkness the dawn peak of activity disappeared but the dusk peak activity was found to persist. However, in constant light these millipedes did not show any rhythmicity. In the present study, we investigated the circadian rhythmicity of the surface-dwelling millipede *Syngalobolus* sp. in DD, LD and LL conditions.

MATERIALS AND METHODS

Adult millipedes *Syngalobolus* sp. were black, with body length of 45.8 ± 2.44 mm and body mass of 93 ± 12 mg, and were collected soon after the monsoon rains during the morning hours. They were held in laboratory vivaria containing a 10 cm layer of moist garden soil covered with algae, grass and decayed wood and leaves as food items. Fresh food was given on alternative days. The animals were maintained at a temperature of $26.8 \pm 0.8^\circ\text{C}$ and 85.8 ± 7.5 % relative humidity. Experiments were carried out under LD (12:12 h), with lights-off at 18.00 h, and in constant darkness and continuous light. The illumination intensity was 10–15 lux.

Recording techniques: The locomotor activity was recorded for 12 millipedes. Each of them was housed in an oval shaped plexiglass chamber, which had a one cm wide path for each of them to move around freely. The movements were picked up by infrared (IR; Pati et al., 1998) sensors, responding to the IR emitter (TIL, 32), which activated the appropriate pens of an Esterline Angus Event Recorder (A 620X) via the amplifiers (fabricated by USIC, M.K. University). The activity patterns over 24 h periods were pasted one below another in chronological order, double plotted to facilitate visualization of data. A torch with red filter (> 650 nm) was used for feeding and cleaning purposes. Phase angle difference (ψ) is defined as the difference in time between the activity onset and the time of lights-off in the LD cycles. If the onset of activity followed lights-off, ψ was taken as -ve and if it anticipated lights-off it was taken as +ve.

RESULTS

All of these surface-dwelling millipedes ($n = 11$) entrained their locomotor activity rhythm to LD (12:12 h) cycles. The activity bouts were practically confined to

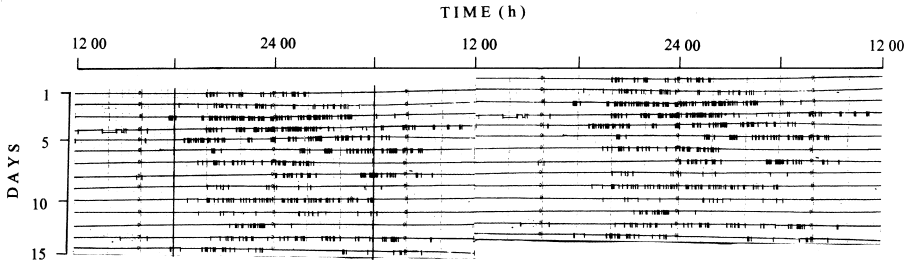


Fig. 1. Entrainment activity pattern of a surface-dwelling millipede *Syngalobolus* sp. under LD (12:12 h) condition. Vertical box indicates period of darkness.

the phase of the darkness (Fig. 1). However, the onset of activity occurred always after the lights went off. Table 1 shows the values for different circadian parameters under different experimental conditions. All specimens showed a negative phase angle difference with values of -83.2 ± 24.72 min. The mean duration of activity of millipedes were 7.37 ± 1.56 h, 7.88 ± 1.03 h and 8.44 ± 0.93 h respectively for LD, DD and LL conditions. Under DD the millipedes freeran with clear-cut activity patterns (Fig. 2). The mean period length was 23.8 ± 1.0 h (range 22.8 to 25.0 h, $n = 9$). Under LL condition the locomotor rhythm persisted (Fig. 3). The period length seemed to be longer than that in DD because in LL it was 25.2 ± 0.1 h (range 25.1 to 25.3 h, $n = 3$).

TABLE 1. Various circadian parameters of surface-dwelling millipede (*Syngalobolus* sp.) under different experimental conditions.

Animal No.	Period length (h)		Phase angle Difference	Duration of activity (h)		
	DD	LL	-ve (min)	LD	DD	LL
1.	23.87 ± 1.6	—	52.5 ± 52.5	6.7 ± 3.46	9.32 ± 3.99	—
2.	25.0 ± 1.13	—	82 ± 57	6.12 ± 3.92	8.15 ± 2.02	—
3.	24.35 ± 0.6	—	89 ± 50	7.6 ± 4.25	8.21 ± 3.17	—
4.	23.27 ± 1.26	—	98 ± 88	7.45 ± 3.85	7.96 ± 3.06	—
5.	22.76 ± 0.73	25.16 ± 0.7	74 ± 68	10.72 ± 6.33	6.77 ± 1.94	7.56 ± 3.6
6.	23.02 ± 1.0	25.12 ± 0.51	102 ± 89	7.27 ± 3.92	9.14 ± 2.19	9.43 ± 3.74
7.	23.0 ± 1.0	25.26 ± 1.3	48 ± 18	9.18 ± 4.6	8.16 ± 1.69	8.35 ± 3.98
8.	25.0 ± 0.52	—	108 ± 46	8.17 ± 4.5	6.41 ± 3.35	—
9.	—	—	46 ± 40	5.98 ± 3.85	—	—
10.	—	—	110 ± 76	5.15 ± 3.88	—	—
11.	—	—	106 ± 56	6.73 ± 3.78	—	—
12.	23.8 ± 0.9	—	—	—	6.8 ± 2.61	—

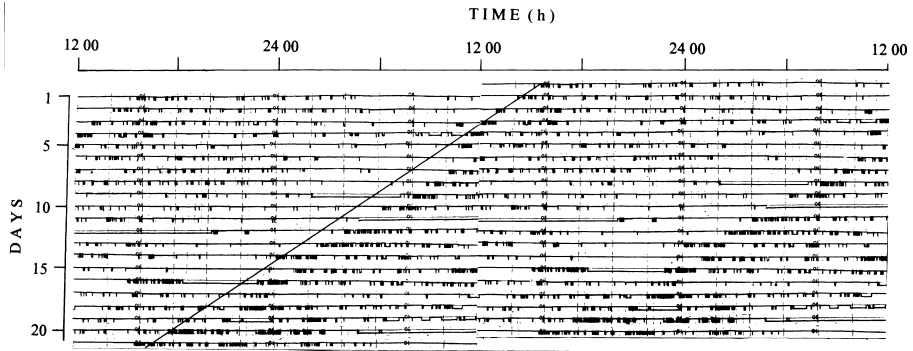


Fig. 2. The freerunning locomotor activity of the millipede *Syngalobolus* sp. recorded under DD condition. The period length is 23.0 h.

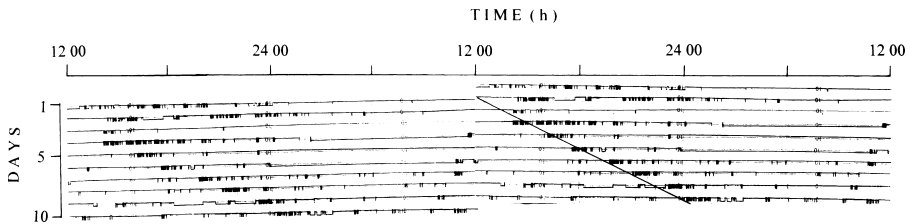


Fig. 3. The freerunning activity pattern of the millipede *Syngalobolus* sp. recorded under LL condition. The period length is 25.3 h.

DISCUSSION

From behavioural observations, Bano and Krishnamoorthy (1979) reported that the locomotor activity of field millipedes peaked during the light phase of the LD (12:12 h) cycle. On the other hand, several authors have stated that field millipedes are nocturnal (Hopkin & Read, 1992), and that in nature about 90% of the millipede populations are active between 22.00 h and 04.00 h (Dondale et al., 1972). Cloudsley-Thompson (1951) reported the existence of the circadian rhythm in activity of the millipede *Ophistreptus* sp. under constant temperature in the laboratory. The results of our experiments with monitoring of the locomotor activity rhythm in the surface dwelling millipede *Syngalobolus* sp. unambiguously showed that the activity occurred during the dark hours and that the rhythmicity persisted under DD and LL and also that this rhythm was entrainable to light-dark cycles. Although in LL we have data only for a few animals ($n = 3$), it appears that

in this species of millipede τ_{LL} was greater than τ_{DD} , thus would be in accordance with the Aschoff's rule (Aschoff, 1960). However, there was no observable difference in the duration of activity in the three different light regimes (LD, DD, and LL).

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