

Arrhythmic perch hopping and rhythmic feeding of starlings in constant light: Separate circadian oscillators?

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Summary. Perch hopping activity and food intake were recorded in starlings in different intensities of continuous illumination (LL), varying from 0.1 to 1000 lux. Circadian rhythmicity in perch hopping disappeared in 10 lux and all higher intensities. In contrast, freerunning circadian rhythms in feeding were always present. In low light intensities, the perch hopping rhythm usually phase leads feeding, increasingly so with shorter circadian period. Locomotor activity may reflect motivational states unrelated to feeding.

Introduction

It is a general experience that freerunning rhythms disappear in constant bright light. Both diurnal and nocturnal organisms – which otherwise have different responses to light (Aschoff 1964) – develop circadian arrhythmicity in conditions of continuous high light intensity. This has been documented for instance for the pupal eclosion rhythm of *Drosophila pseudoobscura* (Pittendrigh 1981) and the rhythms of locomotor activity in diurnal songbirds (McMillan et al. 1975; Binkley 1977; Wever 1980) nocturnal golden hamsters (Pittendrigh and Daan 1976) and rats (Terman and Terman 1980). Similar effects in other species have presumably remained unpublished, as researchers rapidly chose lower light intensities to study their circadian systems in freerun. Yet the incidence of arrhythmicity in constant light may reveal interesting aspects of circadian organization and its relationship with photic sensory input from the environment. Thus McMillan et al. (1975) were able to demonstrate that the arrhythmicity response is mediated through the eyes of house sparrows, in contrast with other photic responses of the circadian system (entrainment, period change and photoperiodic in-

duction) that do not require the presence of the eyes. Arrhythmicity can be interpreted as the absence of circadian oscillations in a central pacemaker. Then, any rhythm in behaviour or physiology governed by the pacemaker will be abolished, as has been suggested for the disappearance of locomotor activity rhythms in pinealectomized house sparrows (Gaston and Menaker 1968). Alternatively, it might be explained as an effect of light on the pathway between central pacemaker and the overt rhythm studied. In this case rhythms in other behavioural or physiological functions are not necessarily abolished, since light may affect pathways differentially. We have studied the effects of constant light of various intensities on the circadian rhythms of feeding and locomotor activity in starlings, and report that the feeding rhythm persists while the perch hopping rhythm disappears.

Materials and methods

Male starlings (*Sturnus vulgaris*) were caught near the laboratory, and kept individually in cages (50 × 50 × 50 cm) visually separated in a temperature controlled (18 ± 2° C) and sound attenuated chamber. Each cage contained two perches one of which was mounted on a microswitch, and a feeder with food pellets (Trouvit®, Fa. Trouw). Access to the food was through a hole (φ 2 cm). Feeding birds interrupted an infrared light beam, aligned with a photocell mounted across the hole. Perch hops and feeding activity were recorded on an event recorder (Esterline Angus, chart speed 1 inch/h) outside the chamber.

Eight fluorescent tubes mounted above the cages provided continuous illumination. Light intensity was controlled by varying the number of powered tubes and by using filters. Light intensities were measured in Lux at the perch levels in the cages in each condition. In a pilot experiment two birds were released from LD 12:12 to LL of unknown high light intensity (>100 Lux). Subsequently, in the main experiment 5 birds were studied for 9 months in various intensities (see Table 1). Three birds lived in good health throughout the experiment. Two died after 8 months due to an accidental obstruction of their food hoppers and were replaced by new birds.

Data were analysed by visual inspection of actograms (see Figs. 1, 2) and by periodogram analysis (Enright 1965) of sam-

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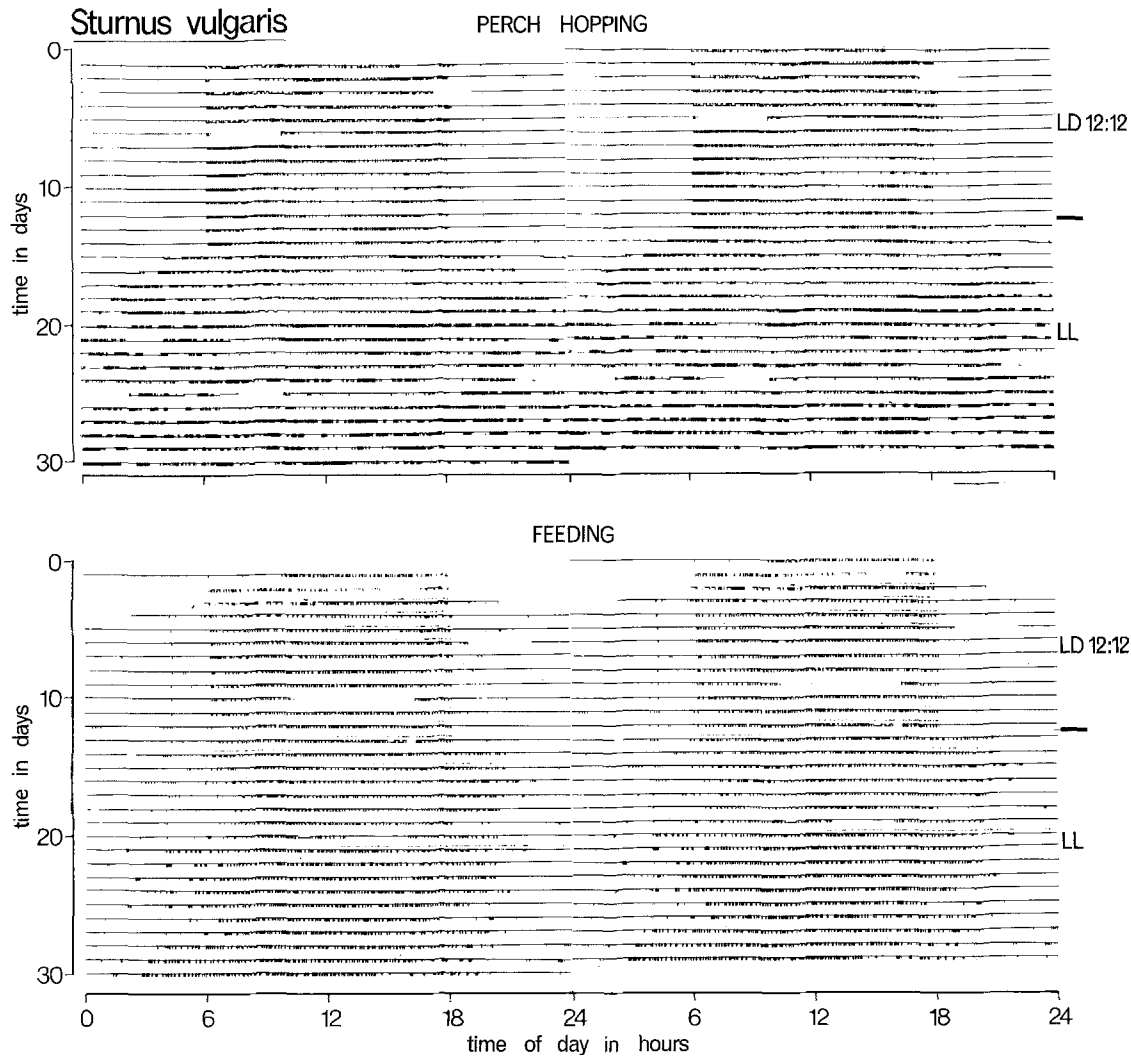


Fig. 1. Simultaneous records of perch hopping (above) and feeding (below) of a starling in LD 12:12 (lights on 6:00–18:00) and, subsequently LL (>100 Lux). The records are double plotted. From day 19 till 30, the circadian perch hopping rhythm has disappeared, while the feeding rhythm remains

Table 1. a Rhythmic status of the birds in a sequence of light intensities. *N* number of birds; *P* number of birds with perch hopping rhythm; *F* number of birds with feeding rhythm. **b** Circadian periods of feeding rhythms (τ_f) and perch hopping rhythms (τ_p).

Exposure (days)	Light intensity (Lux)	a			b	
		<i>N</i>	<i>P</i>	<i>F</i>	$\tau_f \pm \text{SD}$ (h)	$\tau_p \pm \text{SD}$ (h)
43	100	5	0	5	23.63 ± 0.43	—
56	1	5	5	5	23.91 ± 0.35	23.83 ± 0.35
17	0.1	4	4	4	23.99 ± 0.25	23.91 ± 0.53
41	1	5	5	5	23.75 ± 0.25	23.65 ± 0.29
43	10	5	0	5	23.85 ± 0.40	—
39	50	3	0	3	23.96 ± 0.49	—
29	100	4	0	4	23.81 ± 0.36	—
28	1000	5	0	5	23.59 ± 0.43	—
20	50	5	1	5	24.04 ± 0.76	—
16	1	5	4	5	24.01 ± 0.16	24.03 ± 0.26

ple sections of the records. For all birds sections were taken from the final ten days in each light intensity. To quantify the records for periodogram construction each 6 min interval was assigned the value 1 if activity occurred and 0 if no activity occurred. This procedure was checked against a continuous quantitative method using a device which automatically recorded counts of contacts every 20 min. This device was available only during the last month of the study. It produced essentially the same results as the semi-quantitative periodograms (τ -values derived with the two methods for bird 45: feeding in 1000 Lux: 23.9; 23.8 h; feeding in 50 Lux: 24.7; 24.4 h activity in 50 Lux: 24.7; 24.8 h for semi-quantitative and quantitative periodograms, respectively). We have used periodograms for establishing presence or absence of circadian components in the record. When a rhythm was present, we estimated its period (τ) by eyefitting lines through activity onsets over 10-day segments of the record.

Results

The two birds in the pilot experiment showed locomotor activity and food intake only during the

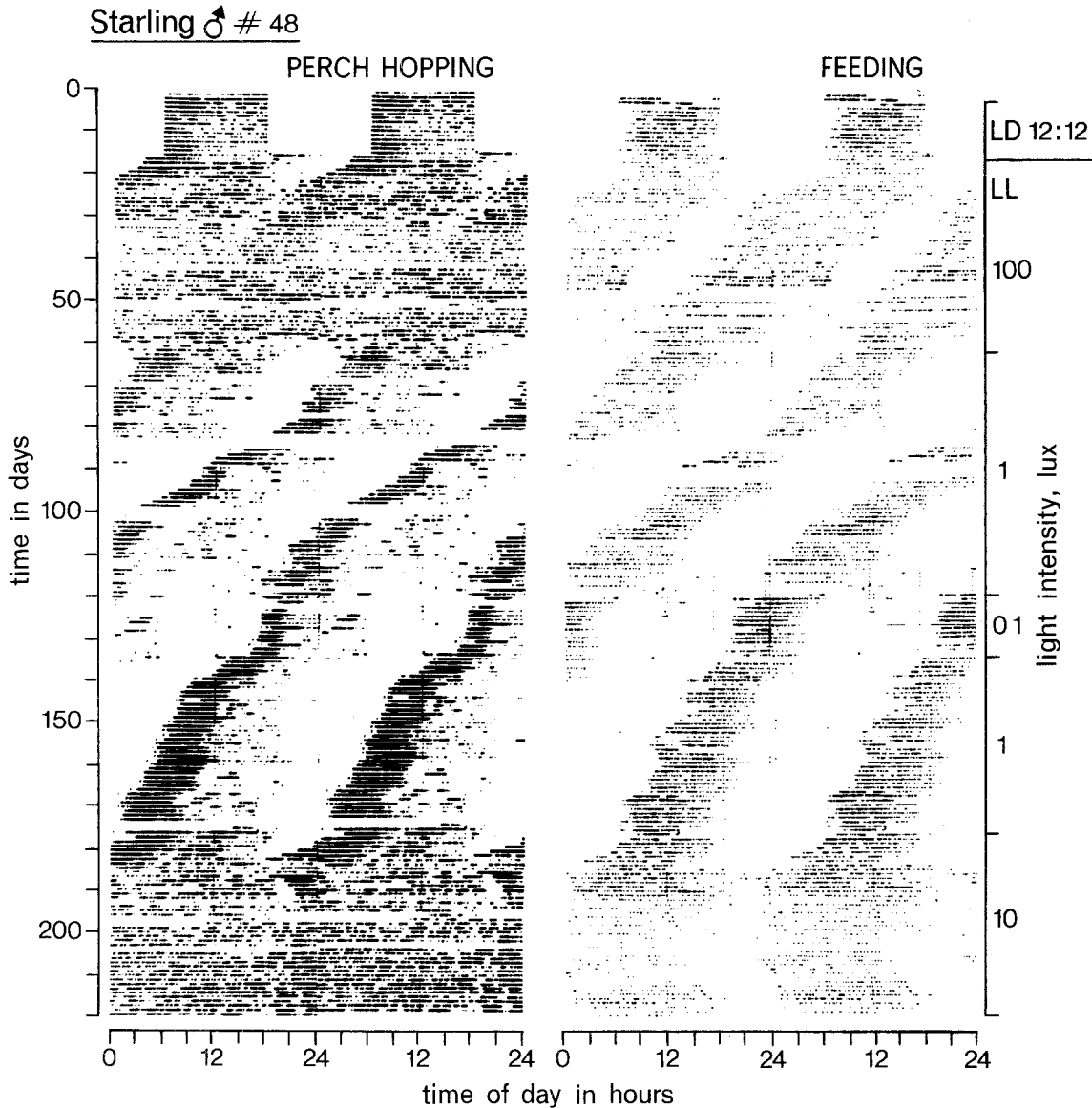


Fig. 2. Simultaneous records of perch hopping (left) and feeding (right) of a starling in LD 12:12 followed by LL of different intensities

light portion of the LD-cycle (Fig. 1). Release in constant light led to a gradual forward shift of the onset of perch hopping until the circadian rhythm had disappeared after about 6 days. In contrast a free running circadian rhythm of feeding remained clearly distinct throughout 18 days of constant light.

In the main experiment both perch hopping and feeding exhibited freerunning circadian rhythms in 0.1 and 1 Lux (Table 1a). However, at higher light intensities perch hopping rhythms disappeared in 26 out of 27 cases whereas simultaneously measured feeding rhythms remained. A representative record (Fig. 2) illustrates this phenomenon. The periodograms for another bird are shown in Fig. 3: there was no light intensity at which the feeding rhythm disappeared. However,

a circadian peak in the perch hopping periodogram vanished when illumination was raised to 10 Lux. It is curious that when returning from high to low intensity the peak returned at 50 Lux in this bird. This is reminiscent of the hysteresis described for light intensity effects in other vertebrate systems, in the incidence of both splitting (Hoffmann 1971) and arrhythmicity (Wever 1980).

Mean circadian periods of the persisting rhythm of food intake were shortest in the highest light intensities (Table 1b). This is in accordance with Aschoff's rule (Aschoff 1960), which was indeed earlier confirmed for starling perch hopping activity (Hoffmann 1960). Perch hopping had a considerable phase lead relative to feeding when both rhythms were present at low light intensities (Fig. 4). The onset of perch hopping sometimes

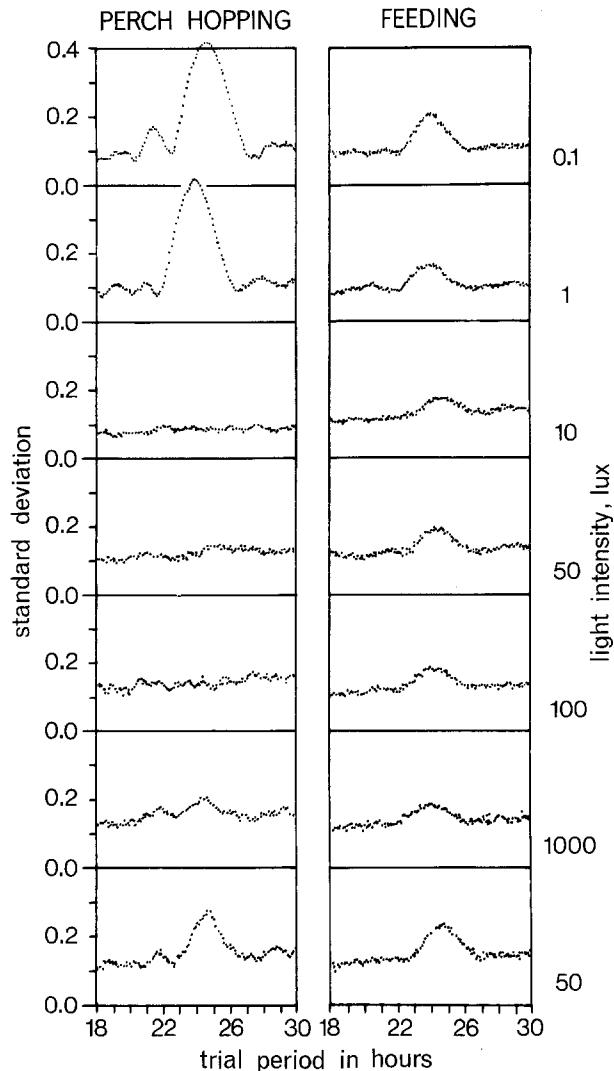


Fig. 3. Periodograms of simultaneous records of perch hopping (left) and feeding (right) of a starling in LL of different intensities

preceded the onset of feeding by as much as 7 h, while the midpoint of perch hopping varied from 4 h before to 3 h after the midpoint of feeding. This phase angle difference (ψ) was negatively correlated with the period (τ) of the perch hopping rhythm (Fig. 4). The difference in phase (and pattern) of the two rhythms was most dramatically illustrated by the record of bird nr. 47 in 1 Lux (Fig. 5). This bird had strongly bimodal perch hopping activity reminiscent of the split rhythms which can be induced in starlings by testosterone injections (Gwinner 1974). The feeding activity was unimodal and occurred in the interval between the two perch hopping bouts rather than during these (Fig. 5).

To illustrate the relevance of the temporal disparity between locomotor activity and feeding under more natural conditions we present data for the daily onset and end of both behaviours in two

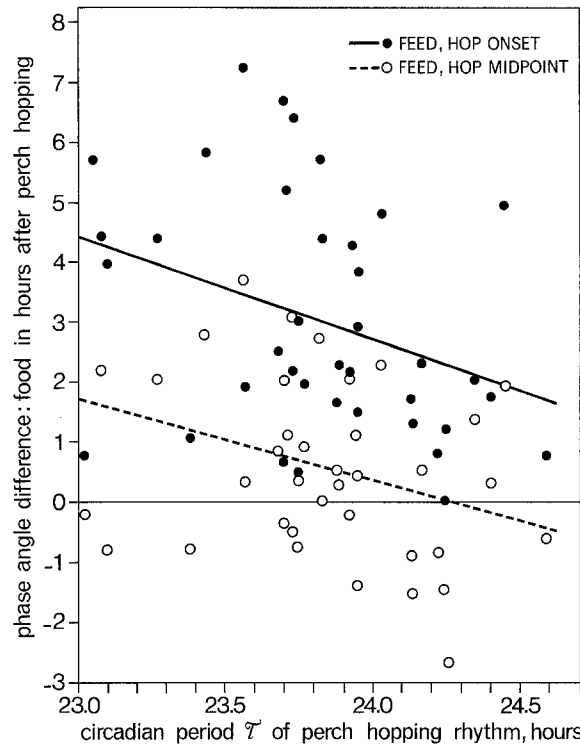


Fig. 4. Phase angle differences between feeding and perch hopping in starlings in LL (0.1 and 1 Lux) as a function of the circadian period of the perch hopping rhythm. Phase angle differences measured either from onsets of feeding and hopping ($n=44.2-1.73$, $r=-0.344$, $n=38$, $P<0.05$) or from midpoints of feeding and hopping (regression $m=31.6-1.30$, $r=-0.341$, $n=38$, $P<0.05$)

birds kept in natural daylight for half a year (Fig. 6). In winter feeding and perch hopping started shortly after dawn civil twilight and stopped just before dusk. In springtime, when daylight increased, the start of feeding was delayed for up to 3 h after the onset of locomotor activity.

Discussion

The results of the main experiment throughout confirmed those of the initial pilot study (Fig. 1). The circadian rhythm of food intake in starlings persisted in constant illumination of intensities high enough to abolish the perch hopping rhythm. This phenomenon inspires the following considerations on the organization of circadian rhythms in birds.

If there is a single circadian pacemaker in birds, arrhythmia in one behavioural output is apparently no indication for arrhythmia of the pacemaker, since other rhythms may still persist. Under this assumption high light intensity would abolish rhythmicity by directly affecting the overt behaviour. In the light of this hypothesis the finding that arrhythmia in sparrows requires retinal photic input whereas other circadian responses to light do not (McMillan et al. 1975) gains new implica-

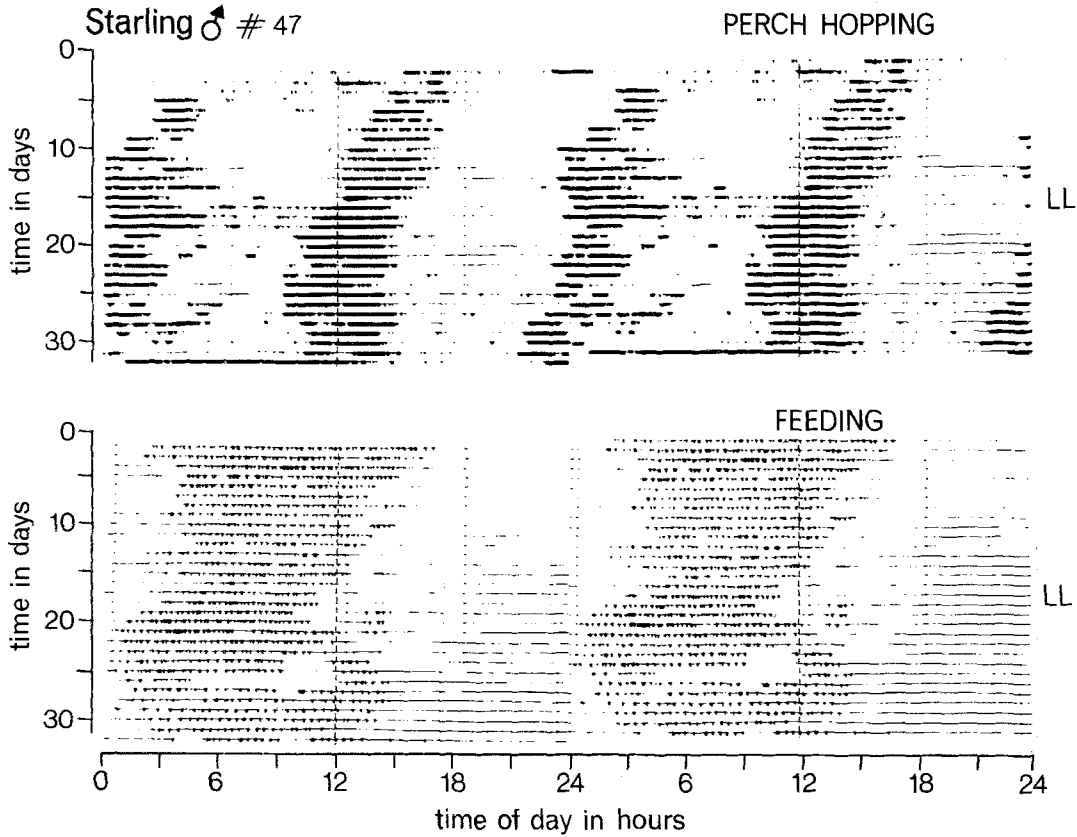


Fig. 5. Simultaneous records of perch hopping (above) and feeding (below) of a starling in LL (1 Lux). Note the difference in circadian distribution of the two behaviours

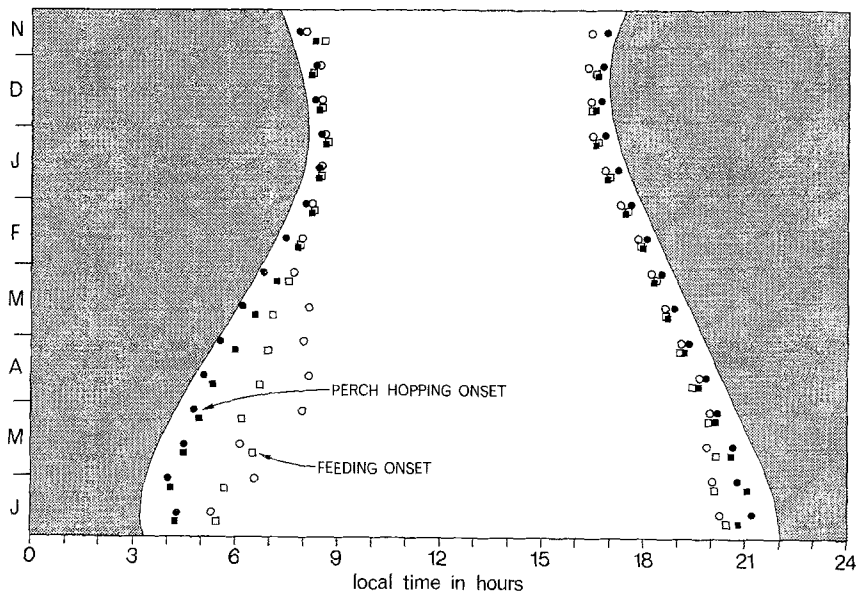


Fig. 6. Mean daily onsets and offsets of perch hopping (solid symbols) and feeding (open symbols) in two starlings in natural daylight. Note the difference in onset in spring

tions. This result would be consistent with the interpretation that the eyes transduce the influence of light on locomotor activity, bypassing the central pacemaker. Experiments testing whether masking effects (Aschoff et al. 1982) of light require retinal input have not been performed.

There may, however, be two endogenous circadian

oscillators, one responsible for perch hopping activity, one for feeding. This is indeed suggested (at low light intensities) by the change in phase relationship between the two with changing τ (Fig. 4). The data do not distinguish between a system of two slave oscillators with different properties but depending on the same pacemaker and

a system with two separate pacemakers. When both oscillators are manifest they run synchronously and hence they are apparently coupled to each other. At least the feeding oscillator has access to light since the period of the feeding rhythm changes with light intensity also in the absence of a circadian locomotor activity rhythm. Increased phase lag of the feeding oscillation with shorter τ suggests that this oscillation is less variable or more stable than the activity oscillator. Similar evidence was presented by Aschoff et al. (1973) for different activity measures (cage movements and wheel running) in hamsters.

The separate control of food intake and locomotor activity rhythms has also implications in a more functional context for our understanding of spontaneous activity. The temporal separation (Figs. 5 and 6) suggests that perch hopping reflects a motivational state unrelated to foraging. Also during LL-induced arrhythmia there are daily episodes of activity without any tendency to feed. This contrasts with the notion of circadian activity as the endogenous basis for periodic food intake. In nature times of activity correspond roughly to times of foraging, except at night in nocturnal migrants (Gwinner 1975). However, there is no precise correspondence if we consider these behaviours quantitatively, and the phasing of the two to the light-dark cycle can be considerably different (Fig. 6).

The difference in timing suggests difference in function. The functional meaning of spontaneous locomotor activity is not thoroughly understood (for a valuable review see Aschoff 1962). The initial period of daily activity in songbirds is usually devoted to territorial advertisement (the 'dawn chorus') and it has been shown that this activity reduces the risk of territorial intrusion around dawn (Kacelnik and Krebs 1983). Perch hopping in cages could be associated with similar motivational states as underly dawn song activity. If perch hopping activity indeed reflects social behaviour, a strong entraining response to light seems ideally suited to keep a precise and stable phase relationship of the onset of activity to sunrise – and to conspecifics. The feeding rhythm of an organism seems more adequate if it can be adjusted to varying times of day when optimal feeding decisions are to be made (Daan 1981) while light has no dominating influence. It may therefore be an adaptive feature of circadian organization that the endogenous circadian oscillation of food intake is less affected by light than the activity rhythm.

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