Activity Rhythms and Masking Response in the Diurnal Fat Sand Rat Under Laboratory Conditions

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Daily rhythms are heavily influenced by light in two major ways. One is through photic entrainment of a circadian clock, and the other is through a more direct process, referred to as masking. Whereas entraining effects of photic stimuli are quite similar in nocturnal and diurnal species, masking is very different. Laboratory conditions differ greatly from what is experienced by individuals in their natural habitat, and several studies have shown that activity patterns can greatly differ between laboratory environment and natural condition. This is especially prevalent in diurnal rodents. We studied the daily rhythms and masking response in the fat sand rat (Psammomys obesus), a diurnal desert rodent, and activity rhythms of Tristram’s jird (Meriones tristrami), a nocturnal member of the same subfamily (Gerbillinae). We found that most sand rats kept on a 12 h:12 h light-dark (LD) cycles at two light intensities (500 and 1000 lux) have a nocturnal phase preferences of general activity and higher body temperature during the dark phase. In most individuals, activity was not as stable that of the nocturnal Tritram’s jirds, which showed a clear and stable nocturnal activity pattern under the same conditions. Sand rats responded to a 6-h phase advance and 6-h phase delay as expected, and, under constant conditions, all tested animals free ran. In contrast with the nocturnal phase preference, fat sand rats did not show a masking response to light pulses during the dark phase or to a dark pulse during the light phase. They did, however, have a significant preference to the light phase under a 3.5 h:3.5 h LD schedule. Currently, we could not identify the underlying mechanisms responsible for the temporal niche switch in this species. However, our results provide us with a valuable tool for further studies of the circadian system of diurnal species, and will hopefully lead us to understanding diurnality, its mechanisms, causes, and consequences.

Keywords: Activity pattern, circadian rhythms, diurnality, meriones tristrami, psammomys obesus

INTRODUCTION

One of the most fundamental features of a species is its activity pattern. It is determined by the interaction between an internal clock that entrains to the environmental periodic changes of the 24-h cycle, and the direct effect of environmental factors on activity. Animals can be characterized as diurnal (day active), nocturnal (night active), crepuscular (twilight active), or cathemeral (arrhythmic), based on the distribution of their locomotor activity throughout the diel cycle. These activity patterns are accompanied by anatomical, physiological, and behavioral adaptations to the specific temporal niche, which further confine the species to its activity phase (Hut et al., 2012; Kronfeld-Schor & Dayan, 2003, 2008).

Daily rhythms are heavily influenced by light in two major ways. One is through photic entrainment of a circadian clock, which is located in the suprachiasmatic nucleus (SCN) in mammals (Moore-Ede et al., 1982), and the other is through a more direct process, referred to as masking, in which light and dark can directly increase or decrease activity (Aschoff, 1960; Aschoff & Vongoetz, 1988; Redlin et al., 2005). Whereas entraining effects of photic stimuli are quite similar in nocturnal and diurnal species (Challet, 2007; Kas & Edgar, 2000; Lee & Labyak, 1997), masking is very different. Specifically, light increases activity in diurnal mammals (positive masking) and suppresses it in nocturnal ones (negative masking), whereas darkness acts in the opposite ways (Aschoff, 1960; Hagenauer & Lee, 2008; Mrosovsky & Hattar, 2003; Mrosovsky et al., 2005; Redlin & Mrosovsky, 2004).

When studying any trait, it is always important to bear in mind that similar phenotypes may result from different selection pressures, and have different underlying mechanisms. The earliest mammals were nocturnal insectivores (Crompton, 1980). Although most current mammals are still nocturnal, independent evolutionary transitions to a diurnal pattern of adaptation...
to the day-night cycle have occurred within both closely and distantly related taxa (Roll et al., 2006). Therefore, the neural mechanisms sustaining diurnality may be diverse (Smale et al., 2003, 2008), and information regarding the circadian system of several different diurnal species is essential for the development of our understanding of the mechanisms regulating activity patterns.

The mammalian circadian system is usually studied under controlled laboratory settings, which enables controlling many environmental variables and creating settings that allow testing the effect of certain variables and hypotheses (Calisi & Bentley, 2009). To study phase preferences, the animals are kept under constant conditions and 12 h:12 h light-dark (LD) cycle, and in order to test if the perceived rhythms are truly circadian, the animal is transferred to constant dark or light conditions (DD or LL). These conditions differ greatly from what is experienced by individuals in their natural habitat; in the context of circadian rhythms research, nature provides a much richer cycling environments, including both biotic (e.g., light, temperature, humidity, and radiation) and abiotic (e.g., predators, prey, competitors, food availability, etc.) cycling variables. Several studies have shown that activity patterns can greatly differ between laboratory environment and natural conditions (reviewed by Calisi & Bentley, 2009; Hut et al., 2012). This is especially prevalent in diurnal rodents. During the past few years, several cases of diurnal rodents switching to nocturnal activity patterns in the laboratory were reported (reviewed by Hut et al., 2012). For example, Degus (*Octodon degu*) are diurnal in their natural habitat (Fulk, 1976), but in the laboratory are able to invert their activity to nocturnal when a running wheel is available (Hagenauer & Lee, 2008; Kas & Edgar, 1999). In a similar way, the unstriped Nile rats (*Arvicanthis niloticus*) are diurnal in the field but are both diurnal and nocturnal in the laboratory (Blanchong & Smale, 2000; Blanchong et al., 1999). Golden spiny mice (*Acomys russatus*) are diurnal in the field but are primarily nocturnal under laboratory conditions (Cohen & Kronfeld-Schor, 2006; Cohen et al., 2009, 2010; Kronfeld et al., 1994; Levy et al., 2007), and a recent study reported that the subterranean rodent tuco-tuco (*Ctenomys aff. Knighti*) emerges above ground during daylight in the field, but in the laboratory its general and wheel-running activity is concentrated in the dark phase (Tomotani et al., 2012). To date, the only studied diurnal rodents that remain strictly diurnal under laboratory conditions are ground squirrels (Everts et al., 2004; Hut et al., 1999a, 1999b).

Several groups have looked at masking in diurnal rodents. Our studies of masking in golden and common spiny mice (*Acomys russatus* and *A. cahirinus*, respectively) have revealed patterns that were highly predictable in some ways but quite novel in others (Cohen et al., 2010; Rotics et al., 2011b). Common spiny mice behaved exactly as expected for a nocturnal rodent, indicating that traits associated with masking do not distinguish the genus *Acomys* from other mammalian taxa. Golden spiny mice increased their activity in response to a dark pulse during the day, as is typical for nocturnal rodents, but there was extreme inter- and intraindividual variability, and did not exhibit significant response to light pulses at night. Moreover, in a 3.5:3.5 h LD cycle golden spiny mice showed a pronounced circadian rhythm but no evidence of masking. Mongolian gerbils (*Meriones unguiculatus*) responded in some ways as expected from nocturnal rodents or did not respond at all: light pulse during the dark phase nearly completely suppressed wheel running activity but had no effect on general activity, whereas dark pulse during the light phase induced wheel running and had no effect on general activity (Weinert et al., 2007). Degu (*Octodon degus*) presents both nocturnal and diurnal chronotypes when given access to a running wheel. In accord with their chronotype, diurnal degus increase their activity in response to a light pulse during the dark phase, whereas in the nocturnal individuals it resulted in a sharp decrease in wheel-running activity (Vivanco et al., 2010). Similarly, Nile grass rats (*Arvicanthis niloticus*) increase nocturnal activity when provided with a running wheel, and their response to light is in accord with their activity pattern: when they are nocturnally active (in the presence of running wheel), activity is suppressed in response to light, whereas in animals without running wheel, light was associated with increased general activity (Redlin & Mrosovsky, 2004). In another study, diurnal Nile grass rats (with no access to a running wheel) increased general activity in response to a light pulse during the dark phase, did not respond to a dark pulse during the light phase, and under a 3.5:3.5 h LD cycle showed a pronounced circadian rhythm and a significant preference to the light phase (Shuboni et al., 2012). From the studies performed to date on diurnal species, it appears that diurnality is indeed a more complex and diverse trait than nocturnality and this raises questions about diurnality, its mechanisms, and stability relative to the rigid activity pattern of nocturnal species. Studying the circadian system of many diurnal species will help us find generalities and understand the mechanisms underlying diurnality.

The fat sand rat (*Psammomys obesus*) is a diurnal desert rodent. In their natural habitat, the Sahara-Arabian desert, sand rats are diurnal (Daly, 1979; Gromov, 2001; Ilan & Yom-Tov, 1990). During summer they show a bimodal activity pattern, avoiding activity during the hot summer mid-day hours, whereas during winter they show a unimodal activity, avoiding the cool mornings and evenings, but in both seasons they are active only during the light hours (Ilan & Yom-Tov, 1990; Mendelssohn & Yom-Tov, 1999). The fat sand rat physiology, including energetics, osmoregulation, and reproduction, was studied extensively in the laboratory (e.g., Degen et al., 1991;
Fichet-Calvet et al., 1999; Haim et al., 2006). Moreover, the fat sand rat is being used in biomedical research, for the study of diabetes mellitus, fatty liver disease, and recently seasonal affective disorder (Ashkenazy et al., 2009a, 2009b; Einat et al., 2006a, 2006b; Khalkhal et al., 2012; King, 2012; Krivisky et al., 2011; Kronfeld-Schor & Einat, 2012; Maislos et al., 2006; Ramachandran et al., 2012). Therefore, many aspects of its physiology are well studied. Surprisingly, relatively little is known about the fat sand rat circadian rhythms. Several laboratory studies reported that rectal body temperature and oxygen consumption of fat sand rats are higher during the light phase (Neuman et al., 2005; Schwimmer et al., 2010). However, its activity pattern has not been described under laboratory conditions, and a study that tested its temporal feeding behavior in the laboratory found no preference to the light or the dark phase, with animals feeding constantly throughout the diel cycle, and no difference in “energy saving” behavior (sleeping and motionless postures) between the diel phases (Khokhlova et al., 2005).

The aim of this work was to study the daily rhythms and masking response in the fat sand rat. To do so, we characterized its body temperature and activity rhythms under various controlled laboratory conditions. As a comparison, we also studied activity rhythms of Tristram’s jird (Meriones tristrami), a nocturnal member of the same subfamily (Gerbillinae) as the fat sand rat (which is actually a gerbil; Scott & Dunstone, 2000). We hypothesized that under 12 h:12 h LD conditions, the sand rats will show a diurnal phase preference, that they will be entrained, showing a free-running rhythm under both LL and DD conditions, and will be able to follow the shift of the LD cycle. In the masking experiments, we hypothesized that the sand rats will respond to the light and dark pulses as expected from a diurnal rodent, i.e., increase activity in response to a light pulse, decrease activity level in response to a dark pulse, and show a preference to the light phase in a 3.5 h:3.5 h LD cycle.

MATERIALS AND METHODS

General
Adult male fat sand rats (approximately 6 mos old, average weight: 174.6 g) purchased from Harlan Laboratories (Jerusalem, Israel) were used in this study. Activity patterns and core body temperature rhythms were studied in the laboratory under controlled conditions (25°C; Ashkenazy et al., 2009a, 2009b; Ashkenazy-Frolinger et al., 2010; Krivisky et al., 2011; Palgi et al., 2005) and daily photoperiod of 12 h:12 h LD (unless specified otherwise). All animals were housed individually in 42 x 26 x 15 cm polycarbonate cage, and due to their susceptibility to develop diabetes they were provided with special low-energy pellets (product 19560; Koplock, Petach-Tikva, Israel) and water ad libitum.

For comparison, we also used five Tristram’s jirds (6–8 mos old, average weight: 72 g) from our breeding colony at the I. Meir Segals Zoological Garden at Tel Aviv University and kept them under the same conditions (25°C, 12 h:12 h LD, 500 lux). General activity was continuously recorded using an infrared detector (Intrusion detector model MH10; Crow group, Kiriat-Teufa, Israel) connected to a personal computer (PC). Data were collected by a PC at 6-min intervals using software designed for this purpose (ICPC, Ntanya, Israel).

All procedures were conducted in accordance with and approved by the Institutional Animal Ethics Committee (L-10-026) and meet the ethical standards of the journal as outlined in Portaluppi et al. (2010). All efforts were made to minimize the number of animals used and their discomfort.

Body Temperature and Activity
Twenty-two sand rats were anesthetized using isoflurane in medical-grade oxygen, using an anesthetic machine (Ohmeda; Louisville, KY, USA), and implanted intraperitoneally with one of two telemetric temperature probes: TA10TA-F40 (Data Sciences, St. Paul, MN, USA; n = 12) or G2 E-Mitter (Mini-Mitter, Bed, OR, USA; n = 10). Skin and abdominal wall were sutured (5-0, Dexon; Syneture, Norwalk, CT, USA), and the incision was treated topically with Silverol Cream (silver sulfadiazine 1%; Teva, Israel). Antibiotics (Baytril 5%, 20 mg/kg; Bayer, Germany) were injected intramuscularly (IM) before implantation, as a prophylactic measure. The cages were placed on top of the receivers for continuous recording of general activity and body temperature. Data were acquired by a Data Quest III collection system (Data Sciences) or by the Vital View data acquisition system (Mini-Mitter).

At least 2 wks of recovery were given to all of the animals before the experiments were started.

Experiment Protocol
In order to allow a comparison with results published for other rodents, we used a typical protocol for studying circadian rhythms: Twelve sand rats were implanted with a TA10TA-F40 transmitter and kept in a 12 h:12 h LD cycle (LD1) in the laboratory. Light intensity during the light phase was approximately 500 lux. After 3 wks, the animals were transferred to constant light (LL) for 4½ wks. At the end of this period, a 12 h:12 h LD (LD2) cycle was reinstated, and after 3 wks the animals were exposed to constant dark (DD). After 4½ wks, a 12 h:12 h LD (LD3) cycle was reinstated for another 3 wks. This was followed by a 6-h phase delay and, 3 wks later, a 6-h phase advance of the LD cycle.

To study the masking responses to light and dark, we kept the 12 sand rats under the same 12 h:12 h LD cycle. After 4 wks, the animals were monitored continuously for 7 d, and on day 8, lights were turned off for 3 h at zeitgeber time (ZT) 2. Seven days after the dark pulse, the same animals were exposed to 3 h of light pulse
started at ZT 14. This was repeated four times using four different light intensities (50, 100, 700, 1500 lux) with 1-wk intervals between each pulse. Two weeks after the last light pulse, animals were exposed to 3.5 h:3.5 h LD cycle and monitored for 2 wks. Only the second week was analyzed.

Since activity and body temperature rhythms that were demonstrated by the animals under 12 h:12 h LD were not consistent with published studies of rectal temperature and oxygen consumption rhythms in this species that were performed under higher illumination (Neuman et al., 2005; Schwimmer et al., 2010, Schwimmer, personal communication), we hypothesized that increasing light intensity will result in diurnal phase preference in the fat sand rat. Therefore, we conducted another experiment under the same conditions but with higher light intensity (1000 lux) using a different monitoring system (G2 E-Mitters; Mini-Mitter) for measuring activity and body temperatures of another 10 animals (Harlan Laboratories) under 12 h:12 h LD cycle for 3 wks.

**Data Analysis**

To determine whether the animals preferred to be active during the light or dark phase under LD cycle, percentage of activity during the light phase was determined as the activity during the 12 h of light divided by the total activity multiplied by 100. Actograms were generated using ClockLab (Actimetrics, Wilmette, IL, USA) and chi-square periodograms were used to determine the significant rhythms (under LL, DD, and 3.5 h:3.5 h).

Statistical tests used included paired t test and repeated-measures one-way analysis of variance (ANOVA) with Scheffe post hoc comparisons. For the light pulses experiment, the data were analyzed as 2 × 4 factorial designs (lighting condition [masking pulse vs. control] × light intensity [1500, 700, 100, 50]) with repeated measures on both factors. SPSS (SPSS, Chicago, IL, USA) was used for all statistical tests. Data are presented as mean ± SEM and p < 0.05 was considered significant.

**RESULTS**

**Sand Rats Under 12 h:12 h LD Cycles**

Average body temperature rhythms (±SEM) of 21 animals (one transmitter malfunctioned) under the two different experimental conditions (500 and 1000 lux) are presented in Figure 1a. In the laboratory, under both light intensities, most of the animals (19 out 21) had significantly higher body temperatures during the dark phase compared with the light phase (1000 lux: t(8) = -2.97, p < 0.05; 500 lux: t(11) = -5.6, p < 0.001; Figure 1b). Body temperature began to rise just before the end of the light phase, and dramatically increased after the lights came off. Average body temperature during the first 2 h of the dark phase was significantly higher than during the light phase and the rest of the dark phase (repeated-measures ANOVA, 1000 lux: F(2,16) = 19.85, p < 0.001; 500 lux: F(2,22) = 25.2, p < 0.001; Figure 1a).

Activity showed the same trend as body temperature under both light intensities (Figures 2a, b, 3): most of the animals displayed a higher percentage of activity during the dark phase. Overall, the percent of general activity during the dark phase was 72.4 ± 2.58% and 68.3 ± 5.6% for the two light intensities, and was significantly lower during the light phase (1000 lux: t(9) = -3.27, p < 0.05; 500 lux: t(11) = -8.65, p < 0.001). Only two animals (e.g., Figure 3f), which also had higher body temperature in the light phase, showed higher percentage of activity (54.96% and 68.53%) during the light phase.

**Tristram’s Jird Under 12 h:1 h LD Cycle**

Figure 4(a–f) depicts actograms of the five tested animals (a–e), and mean (±SEM) percentage activity during the light and dark phases (f). Activity was significantly higher during the dark phase (t(4) = -14.43, p < 0.001, range: 73.77–87.24%). Onset and offset times were defined using the Clocklab program (xx, xx) and were at ZT 11:39 ± 7.8 min and 23:23 ± 8.4 min (ZT ± SEM), respectively.
All fat sand rats \((n = 12)\) displayed free-running rhythms under both LL and DD conditions (e.g., Figure 5); however, other rhythms were also evident and in some individuals activity was scattered across the day. Under LL, the free-running period (FRP) of all individuals except one (male 11) was longer than 24 h \((24.2 \pm 0.22, \text{range} = 23.7–24.6)\). Under DD conditions, the FRP of all individuals was shorter than 24 h \((23.75 \pm 0.15, \text{range} = 23.4–23.9)\), and in all individuals it was shorter than the FRP of the same individual under LL conditions \((t(11) = -6.51, p<0.001)\), except for male 11, for which it was 23.7 under both LL and DD.

Phase Shifts
Since we were unable to define activity onset and offset in most individuals using Clocklab or by visual inspection, we used a clear peak of activity in each individual and studied its adjustment to the phase shift (Cohen & Kronfeld-Schor, 2006; Cohen et al., 2009). Average time of peak activity during the last 10 d before the shift was set to 0, and the changes in the activity peak were described in relation to that time point. In order to define re-entrainment after the shifts, we looked at the first day out of three continuous days following the change, where the activity peak didn’t delay or advanced more than 0.5 h. If a clear activity peak was not seen in an individual sand rat, it was excluded from this analysis (2 of the 12 animals that were studied). As one transmitter ceased at this stage and one individual split its activity in response to the delay (Figure 7b), we compared the rest of the individuals \((n = 8)\; \text{(Figure 6).}\) In these animals, \(6 \pm 2.82 \, \text{d} \; (\text{range} = 2–11)\) were required for re-entrainment after 6-h phase delay and \(11.35 \pm 4.13 \, \text{d} \; (\text{range} = 4–15)\) after 6-h phase advance of the LD cycle; Figures 6, 7a). The number of days required for re-entrainment was almost significantly shorter after the 6-h phase delay compared with the 6-h phase advance of the LD cycle \((\text{paired} \, t \, \text{test,} \; p = 0.051)\).

Dark and Light Pulses

**Dark Pulse**
The response to the 3-h dark pulse at ZT 2 was not the same for all individuals. Eight individuals increased activity level (positive masking), two individuals decreased activity (negative masking), and two did not respond to the dark pulse (activity didn’t change by more than 10%). Average activity of all individuals was not affected by the dark pulse \((5.95 \pm 0.88 \, \text{counts per minute during the dark pulse, compared with an average} 4.5 \pm 0.46 \, \text{counts per minute during the same time during the 7 d before the dark pulse,} \; t(11) = -1.6, \text{nonsignificant [NS]})\).

**Light Pulses**
The responses to 3-h light pulses between ZT 14 and ZT 17 were again not consistent. There was no overall effect of lighting condition \((F(1, 11) = 0.15, \text{NS}), or light intensity \((F(3, 33) = 0.84, \text{NS})\) on activity levels, and no interaction between these variables \((F(3, 33) = 2.05, \text{NS})\).

**3.5 h:3.5 h LD Cycle**
Under 3.5 h:3.5 h LD cycle, most individuals had significant \(~24-\) and \(~7-h\) rhythms, but the robustness of these rhythms varied between individuals (Figure 8). Overall, animals exhibited significantly higher activity level during the light phase of the 3.5 h:3.5 h LD cycle \((56.12 \pm 2.21\%, \; t(11) = 3.46, \; p<0.01)\).

**DISCUSSION**
In this study, we found that, under laboratory conditions, fat sand rats, which are strictly diurnal in the field, have a nocturnal phase preference of general activity and, in accordance, higher body temperature during the dark phase. In most individuals, body temperature and general activity were lowest during the light phase, and a clear peak in activity was always seen right after lights off. We hypothesized that increasing light intensity from 500 to 1000 lux will result in a diurnal phase preference in the fat sand rats. However, average percentage of nocturnal activity was similar under the two light intensities, and activity onsets and offsets were more robust, predictable, and precise under the higher light level (Figure 3). These results are in contrast to physiological rhythms (body temperature and oxygen consumption; Neuman et al., 2005; Schwimmer et al., 2010) reported in this species under similar laboratory conditions.
conditions (food type and availability, cage size, ambient temperature, and light intensities). The main difference we could find is the methods used for measuring rhythms, and the variable measured (e.g., manual measurement of rectal temperature at four time points during the day, or oxygen consumption), but we do not think this is the reason for the different results.

Regardless of the variables that affected activity pattern of the fat sand rats in the laboratory, our results add the fat sand rat to the growing list of diurnal rodents that show a nocturnal phase preference under laboratory conditions (reviewed by Hut et al., 2012).

Even though many of the fat sand rat individuals had a nocturnal phase preference, they were active also

FIGURE 3. Representative double-daily plots of general activity of fat sand rats kept under 12 h:12 h LD conditions, at two different light intensities: 500 lux (a–c) and 1000 lux (d–f). Black bars at top of plot represent the lighting schedule. Individual f was the most diurnal, with 68.53% of its activity occurring during the light phase.
during the day, and cannot be classified as strictly nocturnal. Of the two individuals (out of 21) that exhibited higher percentage of activity during the light phase (54.96% and 68.53%), only one showed stable entrainment to the LD cycle (Figure 3f), whereas the other individual displayed activity that was scattered across the day. In general, regardless of phase preference, activity was not as stable as that of the nocturnal Tritram’s jirds, which showed a clear and stable nocturnal activity pattern, as often reported for other nocturnal species (Refinetti, 2006; Weber & Hohn, 2005). Some form of instability of the rhythms

FIGURE 4. Double-daily plots of general activity in Tristram’s jirds kept under 12 h:12 h LD conditions (a–e) and percent general activity level (average ± SEM) during the light (white) and (dark) phases (f). ***p<0.001.
was described previously in several diurnal rodents (e.g., Blanchong et al., 1999; Cohen & Kronfeld-Schor, 2006; Cohen et al., 2009; Hagenauer & Lee, 2008; Refinetti, 2006; Tomotani et al., 2012), and is less common in nocturnal species, even though most rodents are nocturnal (Roll et al., 2006). In accordance with our current findings, a recent study in our laboratory that compared daily rhythms in anxiety-like behavior in three nocturnal and three diurnal rodent species found that the three nocturnal species (common spiny mouse, rats, and Tristram’s jirds) had significantly lower levels of anxiety during the dark phase, whereas the diurnal species showed either an inverse pattern to that of the nocturnal species (degu) or no rhythm at all (golden spiny mice and fat sand rats; Bilu & Kronfeld-Schor, 2013). These findings may reflect a possible role of the diel changes in anxiety levels in the regulation of behavior and activity throughout the diel cycle: Whereas the nocturnal species all presented a rather similar anxiety-like behavior pattern, with higher anxiety-like behavior during the rest hours, the results for diurnal species showed variation.

In contrast with the nocturnal phase preference of the fat sand rats, the diurnal species did not show a masking response to light pulse during the dark phase or to a dark pulse during the light phase. They did, however, have a significant preference to the light phase under a 3.5 h:3.5 h LD schedule. Several studies of masking response in diurnal rodents compared the same species in the presence and absence of a running wheel, which changes the activity phase preference of the studied species (Otalora et al., 2010; Redlin & Mrosovsky, 2004; Vivanco et al., 2010; Weinert et al., 2007). In these studies, the plasticity in activity pattern occurs within the lifetime of an individual. Other studies (Cohen et al., 2010; Rotics et al., 2011a, 2011b; Shuboni et al., 2012), including the current one, are looking at the evolutionary scale changes that are required for the development of a diurnal phase preference. At this evolutionary scale, the results are inconsistent between different diurnal species, and even between different studies on the same species (e.g., Redlin & Mrosovsky, 2004; Shuboni et al., 2012). Nevertheless, it is clear that some changes occurred during the evolutionary transition from the nocturnal ancestral activity pattern to the

FIGURE 5. Representative double-daily plots of general activity (a) and body temperature (b) of sand rat (no. 9) during 12 h:12 h LD, LL, 12 h:12 h LD, DD, and 12 h:12 h LD. Black bars at top of plot represent the light schedule during the LD periods.

FIGURE 6. A schematic representation of peak activity re-entrainment to the 6-h phase delay and a 6-h phase advance of the 12 h:12 h LD cycles of 8 individuals.
more recent diurnal activity pattern (Roll et al., 2006) of the extant diurnal rodents, allowing the removal of the negative masking effect of light on activity, and that the mechanism underlying diurnality, including the masking response to light and darkness, are diverse (Roll et al., 2006; Smale et al., 2003, 2008).

It seems that although body temperature and activity patterns of the fat sand rat under laboratory conditions are not completely stable and predictable, the animals’ circadian system is functional and synchronized. It responded to a phase advance and phase delay as expected, and under constant conditions, all fat sand rats tested free ran. As previously described in other diurnal mammals (Cohen & Kronfeld-Schor, 2006; Schumann et al., 2005), the effect of constant conditions on the FRP didn’t follow Aschoff’s rule (Aschoff, 1960):

FIGURE 7. Double-daily plots of general activity of sand rats during a 6-h phase delay and a 6-h phase advance of the 12 h:12 h LD cycle. (a) A representative individual that delayed its activity in response to phase delay and advanced its activity in response to the phase advance. (b) an individual that split its activity in response to phase delay and responded to the phase advance but did not entrained to the new LD cycle. Black bars at top of the plot represent the lighting conditions during the normal LD periods.

FIGURE 8. Representative 28-h plots of general activity of sand rats exposed to a 3.5 h:3.5 h LD cycle (a–c) (white background represent the light hours) with corresponding periodograms of the same individual (d–f).
FRP of the fat sand rats were longer than 24 h under LL conditions and shorter than 24 h under DD conditions, as expected from nocturnal animals.

The diet of the fat sand rat consists mainly of chenopods bushes, which are low-quality food, high in fiber, water, and salt and low in nutrients and energy content (Daly, 1979; Degen et al., 1991, 2000; Gromov, 2001; Kam & Degen 1989; Khokhlova et al., 2005; Tchabovsky & Krasno 2002; Tchabovsky et al., 2001a, 2001b). The above-ground activity of the fat sand rats in nature includes both feeding and food caching (Gromov, 2001; Tchabovsky & Krasnov, 2002; Tchabovsky et al., 2001a, 2001b), and it is possible that the large amount of forage required by the fat sand rats to compensate for the low-quality food necessitates consumption of food throughout the light and dark phases, as found in a laboratory experiment (Khokhlova et al., 2005), and that in nature, the food that is hoarded in the borrows during the day is consumed during the night. A similar result of arrhythmia in feeding activity and maintenance of daily activity rhythm was described in another diurnal rodent, the degu (Garcia-Allegue et al., 1999), whose diet include grasses and leaves of shrubs, although they will also take seeds (Bozinovic et al., 2003). Most desert small mammals are granivorous and nocturnal, whereas none of the diurnal desert rodents is granivorous, due to consideration of water balance (Khokhlova et al., 2005). The larger size of the fat sand rat (as well as the degu) may allow it to be folivorous and entails larger thermal inertia (Haim et al., 2006), and both are expected to enable it to be active during the hot desert day. As most desert predators are nocturnal, diurnal activity will result in lower risk of predation, and therefore is advantageous if possible (Khokhlova et al., 2005).

One of the interesting questions that arise from the study of activity patterns of diurnal rodents that switch activity patterns is if temporal niche is easy to be changed, is diurnality still a “trait”? We think it is. The switch from the ancestral nocturnal activity to a diurnal activity pattern requires a set of anatomical, physiological, and behavioral adaptations, including the removal of the negative masking effect of light, and sensory and thermoregulatory capabilities, which occur over the course of evolution (reviewed by Hut et al., 2012; Kronfeld-Schor & Dayan 2003, 2008). A species that is diurnally active in its natural habitat, such as the fat sand rat or the golden spiny mouse, is not a nocturnal species active diurnally—rather, it developed a set of adaptations that support its diurnal activity. For example, the fat sand rat has a remarkably cone-rich retina (41% of total photoreceptor numbers in both central and peripheral retina) that is adapted to daylight vision (Saidi et al., 2011). In our view, such species should be considered diurnal.

Currently, we cannot identify the underlying mechanisms responsible for the temporal niche switch in this species. It is possible that the switch to diurnal activity included reduced robustness or coupling of the SCN clock and peripheral or other (e.g., feeding or Methamphetamine-sensitive circadian oscillator (MASCO); Mohawk et al., 2012) oscillators. In nocturnal rodents, the SCN controls peripheral and other oscillators (e.g., by controlling feeding, body temperature, and hormonal rhythms) and the timing system is hierarchically organized (Mohawk et al., 2012). Diurnal rodents that switch activity to nocturnal under laboratory conditions may require several environmental cycling inputs (apart from the lab LD cycle) for full entrainment of diurnal activity pattern. Nature provides a much richer cycling environments, including both biotic (e.g., light, temperature, humidity, and radiation) and abiotic (e.g., predators, prey, competitors, food availability, etc.) cycling variables. These may entrain not only the SCN, but also peripheral and other oscillators (e.g., feeding-entrainable oscillator), resulting in a diurnal activity pattern. This hypothesis remains to be studied.

DECLARATION OF INTEREST

The authors report no conflicts of interest. The authors alone are responsible for the content and writing of the paper.

REFERENCES


