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Variability of diurnality in laboratory rodents

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Abstract The locomotor activity rhythms of domestic mice, laboratory rats, Syrian hamsters, Siberian hamsters, Mongolian gerbils, degus, and Nile grass rats were compared. Running-wheel activity was monitored under a light–dark cycle with 12 h of light and 12 h of darkness per day. Nile grass rats were found to be reliably diurnal, whereas laboratory rats, Siberian hamsters, domestic mice, and Syrian hamsters were reliably nocturnal. Both diurnal and nocturnal subgroups were observed in Mongolian gerbils and degus. A downward gradient of diurnality was observed from Mongolian gerbils classified as diurnal, degus classified as diurnal, gerbils classified as nocturnal, and degus classified as nocturnal. Nocturnal degus remained nocturnal when tested with an infrared motion detector without running wheels. Thus, although the diurnal–nocturnal dichotomy could be applied to some of the species, it was not appropriate for others. The dichotomy may reflect researchers’ needs for systematization more than a natural distinction between species. Through mechanisms as yet poorly understood, the balance between entraining and masking processes seems to generate a gradient of temporal niches that runs from predominantly diurnal species to predominantly nocturnal species with many chronotypes in between, including species that exhibit wide intra-species gradients of temporal niche.

Keywords Circadian rhythm · Diurnality · Locomotor activity · *Arvicanthis niloticus* · *Meriones unguiculatus* · *Mesocricetus auratus* · *Mus musculus* · *Octodon degus* · *Phodopus sungorus* · *Rattus norvegicus*

Abbreviations LD: Light–dark cycle · SD: Standard deviation

Introduction

The alternation of day and night is an ineluctable geophysical phenomenon that has had a strong impact on life on earth throughout evolution. The choice of a nocturnal or diurnal niche by various life forms was probably guided, at least at first, by a conflict in the exploitation of solar energy, which provided life-sustaining energy to photoautotrophic organisms but also contained highly damaging ultraviolet emissions. At some point, endogenous rhythmicity evolved as a mechanism that allowed organisms to prepare in advance for predictable daily changes in the environment (Engelmann 1988; Halberg 1953; Marques and Waterhouse 1994; Roenneberg and Merrow 2002; Rutter et al. 2002).

Although the difference between extant diurnal and nocturnal organisms may often be quite obvious (that is, diurnal organisms are active mostly during the day and nocturnal organisms are active mostly during the night), very little is known about what makes diurnal organisms be different from nocturnal ones. In animals, eyes specialized for day vision (that is, eyes possessing retinal cones in addition to retinal rods) evidently facilitate adaptation to a diurnal niche, but image-forming photoreception has been shown to be essentially independent from circadian photoreception because the circadian system is responsive to photic stimulation of the eyes even in animals deprived of visual photoreceptors (Foster et al. 1991; Freedman et al. 1999; Goldman et al. 1997; Klerman et al. 2002; Mrosovsky 2003a; Semo et al. 2003; Yoshimura et al. 2002). It seems that the circadian system can acquire information about illuminance levels in the environment both through classical photoreceptors and through photosensitive ganglion cells that use melanopsin as a photopigment (Gooley

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et al. 2003; Hattar et al. 2002; Panda et al. 2002; Ruby et al. 2002a). Researchers who have tried to identify the mechanisms responsible for diurnality or nocturnality have generally found that there is no clear difference between diurnal and nocturnal organisms except for the obvious difference in the phase angles of entrainment—that is, diurnal animals are diurnal because they are active during the day, and nocturnal animals are nocturnal because they are active at night (Dardente et al. 2004; Fidler and Gwinner 2003; Lincoln et al. 2002; Mrosovsky 2003b; Refinetti 1996a; Smale et al. 2003).

Part of the problem in identifying the mechanisms of diurnality or nocturnality may lie on the use of a dichotomous classification. Analysis of data from a multitude of species indicates that the daily rhythm of activity of most animal species is bimodal, meaning that activity is concentrated at sunrise and sunset (Aschoff 1966). This crepuscular pattern of activity is often but not always accompanied by additional activity between dawn and dusk (“diurnal” animals) or between dusk and dawn (“nocturnal” animals). Unfortunately, detailed studies of the variability of diurnality among different species, or even within a single species, have rarely been conducted. The goal of this study was to investigate the variability of diurnality within and between various species of laboratory rodents as assessed through their rhythms of running-wheel activity.

Materials and methods

Subjects

Animals of seven different species served as subjects. The names and characteristics of the species are listed in Table 1. The species included standard laboratory rodents (domestic mouse, laboratory rat, Syrian hamster, Siberian hamster, and Mongolian gerbil) as well as “exotic” rodents (degu and Nile grass rat). All animals were 2–5 months old.

To avoid sampling bias, animals were not prescreened for the quality of their locomotor activity rhythms. However, because very poor performance in the running wheels due to animal idiosyncrasy cannot be reliably distinguished from poor performance due to illness or to equipment malfunction, animals displaying fewer than 100 revolutions per day (the equivalent of a

linear displacement of only 50 m) were not included in the data analysis. There were few such cases (one domestic mouse, one laboratory rat, one degu, and two Nile grass rats), and the sample sizes given in Table 1 do not include these disqualified animals.

Procedure

All animals were housed individually in polypropylene cages (24×36×19 cm) lined with wood shavings and fed Purina rodent chow (Lab Diet 5001) and water ad libitum. A metallic running wheel (12 cm diameter for species under 80 g and 18 cm diameter for larger species) was attached to each animal cage. Magnetic switches attached to the running wheels were connected to data acquisition boards (Digital Input Card AR-B2001, Acrosser Technology, Taiwan). The data acquisition boards were connected to computers that recorded the number of wheel revolutions in 6-min bins (i.e., 0.1 h intervals).

To verify the appropriateness of the use of running wheels for the monitoring of locomotor activity, selected animals were tested both with and without wheels. The animals were housed in transparent polycarbonate cages, and locomotor activity was monitored by an infrared motion detector (Product 49–312, Radio Shack Corp., Fort Worth, TX, USA). The feedback light in the motion detector was disconnected to prevent unintended photic stimulation (Hofstetter et al. 2005), and the output switch terminals were connected to the data acquisition board.

The animal cages were maintained in individual light-tight, ventilated chambers at $24 \pm 2^\circ\text{C}$. Lighting conditions in each chamber were controlled by a programmable electronic timer (ChronTrol XT, ChronTrol Corp., San Diego, CA, USA) that activated white fluorescent bulbs (General Electric F4T5CW) generating an illuminance of approximately 360 lx (range: 340–390 lx across chambers), as measured 8 cm above the cage floor. All animals were housed under a light–dark (LD) cycle with 12 h of light per day (LD 12:12) for 3 or more weeks. After stable entrainment to the LD cycle had been established for each individual, data were collected for 10 days and analyzed for several rhythmic parameters.

After the study was completed, some animals were maintained under the LD cycle for several weeks, some were transferred to constant darkness, and some were

Table 1 Characteristics of the seven species used in the study

Species	Scientific name	Body mass (g)	Sample size	Sex	Source
Degu	<i>Octodon degus</i>	240	25	Mixed	Sandy's Lakeside (Gaffney, SC)
Domestic mouse (C57BL/6)	<i>Mus musculus</i>	30	32	Male	Charles River Laboratories (Wilmington, MA)
Laboratory rat (Wistar)	<i>Rattus norvegicus</i>	360	24	Male	Charles River Laboratories (Wilmington, MA)
Mongolian gerbil	<i>Meriones unguiculatus</i>	60	29	Male	Charles River Laboratories (Wilmington, MA)
Nile grass rat	<i>Arvicantis niloticus</i>	120	32	Mixed	Local colony (see Refinetti 2004d)
Siberian hamster	<i>Phodopus sungorus</i>	45	24	Mixed	Sun Pet Ltd. (Atlanta, GA)
Syrian hamster	<i>Mesocricetus auratus</i>	150	24	Male	Charles River Laboratories (Wilmington, MA)

exposed to an ultradian LD cycle (LD 3.5:3.5), which does not cause entrainment but allows the study of photic masking responses (Redlin and Mrosovsky 1999).

Data analysis

For descriptive purposes, daily wave-form plots of activity were generated for each species by averaging 10 consecutive days for each animal and then averaging all animals in each species (with 6-min resolution).

For each individual of each species, five parameters of the activity rhythm were analyzed: activity level, diurnality, onset time, acrophase, and robustness. The overall activity level was computed simply as the number of wheel revolutions per 24 h. A diurnality index was computed as the ratio of the number of wheel revolutions during the light phase and the number of wheel revolutions during the whole day—i.e., $R_L/(R_L + R_D)$. Animals with indices below 0.5 are more active during the night than during the day (i.e., they are nocturnal), whereas animals with indices above 0.5 are more active during the day than during the night (i.e., they are diurnal).

Onset time (the time of initiation of running activity each day) was determined by a computer algorithm. First, the activity records over 10 consecutive days (2,400 data points) were digitized and educed. That is, time bins containing one or more wheel revolutions were coded as “1”, whereas time bins containing no revolutions were coded as “0”, and the values for the 10 days were added, bin by bin, into a 24-h time series. Next, the algorithm searched for bins whose values were preceded by 2 h with activity below 1.5 times the mean of the time series and followed by 2 h with activity above 1.5 times the mean. If such a bin was located, and if it contained activity on at least 5 of the 10 days, it was considered to be the time of onset. Although complex, this algorithm is very powerful and succeeds in locating the correct onset time (as confirmed by visual inspection of actograms) in all but the noisiest records. In only 2 out of more than 200 data sets did the algorithm fail to identify the onset. Onset times were determined by visual inspection in these two cases.

For computation of the acrophase (the time of the daily peak of the activity rhythm), an iterative curve-fitting procedure based on the single cosinor procedure (Nelson et al. 1979) was used. For each animal, a cosine wave was fitted to the data points according to the function $Y_t = M + A \cos(\theta_t + \phi)$, where Y_t denotes each data point in the time series, M is the mean level of the rhythm, A is the amplitude, θ_t is the trigonometric angle corresponding to time t , and ϕ is the angle displacement for the acrophase. The value of ϕ was determined by iteration: the true value of ϕ was considered to be the one that produced the smallest sum of squares of the deviations between iterated cosine functions and the raw data.

Rhythm robustness (the strength of rhythmicity of the data set) was computed as the Q_P statistic of Sokolove and Bushell's (1978) chi-square periodogram procedure for $P = 24.0$ h. For 10-day-long data sets with 6-min resolution, a 100% rhythmic 24-h pattern (such as a cosine wave) yields a Q_P of 2,400. Rhythm robustness of experimental time series can be reliably determined by comparison of Q_P values with the ideal value of 2,400 (Refinetti 2004b). The chi-square periodogram procedure was also used to calculate the circadian period of animals maintained in constant darkness.

For each of the five parameters, the significance of differences between means of the eight species was tested by analysis of variance (ANOVA) followed by post hoc pairwise comparisons by Tukey's HSD test (Kirk 1995). The level of significance ($\alpha = 0.05$) was maintained at each ANOVA. Variances were compared by pairwise F tests (Kirk 1995). Intra- and inter-species correlation coefficients for various parameters were computed by the principle of least squares (Hays 1988). To avoid inflation of the level of significance due to multiple testing, the significance of correlation coefficients and pairwise F tests was evaluated at $\alpha = 0.001$ so that the overall significance level for all correlations or for all variance comparisons remained close to $\alpha = 0.05$ (Bonferroni correction).

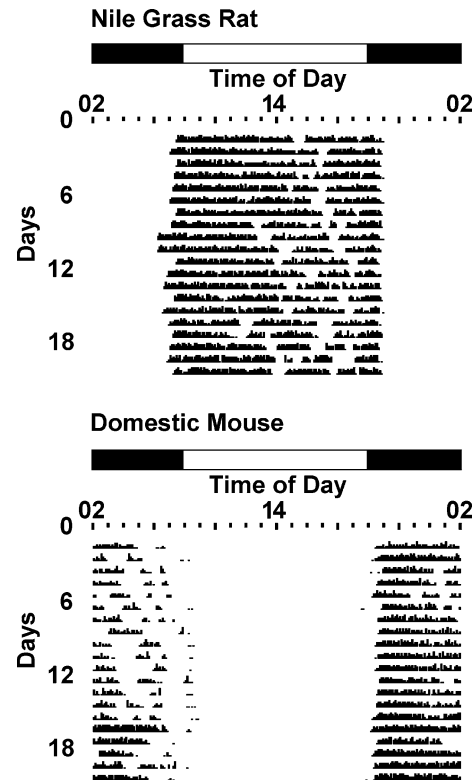


Fig. 1 Actograms of the running-wheel activity rhythms of a representative Nile grass rat and a representative domestic mouse. Time of day is indicated on the horizontal axis and number of days on the vertical axis. The white and black bars above the actograms indicate the duration of the light and dark phases of the light–dark cycle, respectively

Results

Figure 1 shows actograms of the running-wheel activity records of a representative Nile grass rat and a representative domestic mouse. The grass rat initiated activity about 1 h prior to lights-on each day and ran throughout the light phase. The mouse initiated activity at lights-off each day and ran throughout the dark phase. Both animals exhibited brief, irregular intervals of rest during the active phase.

Although most species exhibited activity patterns that were either predominantly diurnal or predominantly nocturnal, Mongolian gerbils and degus were naturally distributed into two distinct groups, as illustrated for degus in Fig. 2. Slightly less than half of the degus ($n=11$) initiated activity about 1 h prior to lights-on each day and ran during most of the light phase and initial hours of the dark phase. The remaining degus ($n=14$) ran mostly during the dark phase, although some exhibited greater activity during the light–dark and dark–light transitions. Four of the degus started out with a clear nocturnal pattern of activity but spontaneously switched to a more diurnal pattern after a few weeks, as exemplified in Fig. 3. Because all animals had been kept under the same LD cycle before arriving in the

laboratory, the switch from nocturnal to diurnal pattern of activity cannot be explained simply by a slow re-entrainment to the laboratory’s LD cycle. All 14 degus reported as nocturnal here were still night-active 3 months after the beginning of the study. No switches from a diurnal pattern to a nocturnal pattern were observed. No switches in either direction were observed in Mongolian gerbils, which were consistently diurnal ($n=22$) or nocturnal ($n=7$).

Figure 4 shows wave-form plots of activity that were generated for each species by averaging 10 consecutive days for each animal and then averaging all animals in

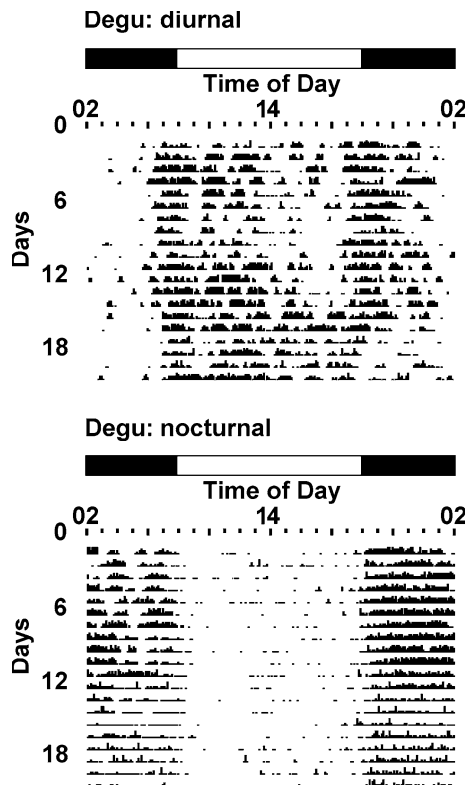


Fig. 2 Actograms of the running-wheel activity rhythms of a diurnal and a nocturnal degu. Time of day is indicated on the horizontal axis and number of days on the vertical axis. The white and black bars above the actograms indicate the duration of the light and dark phases of the light–dark cycle, respectively

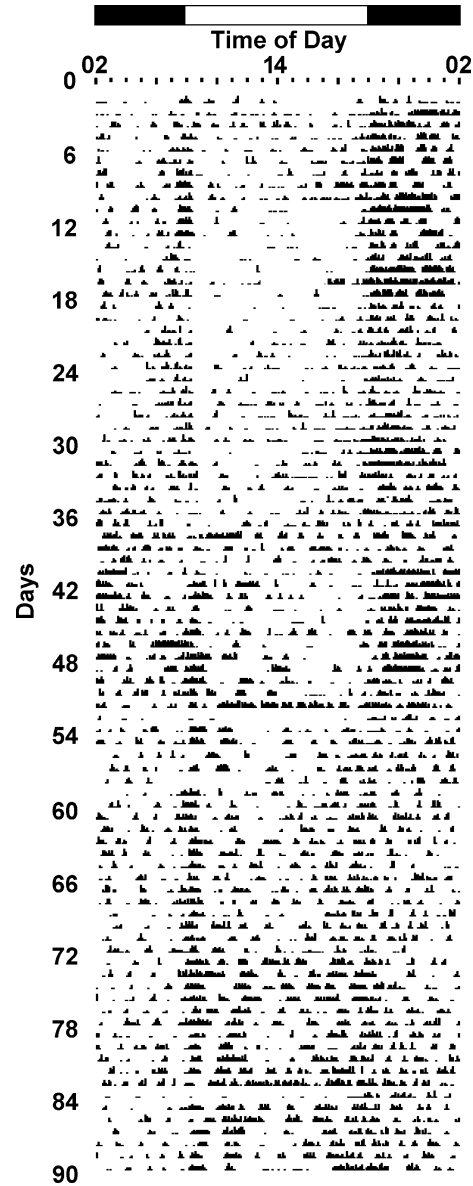


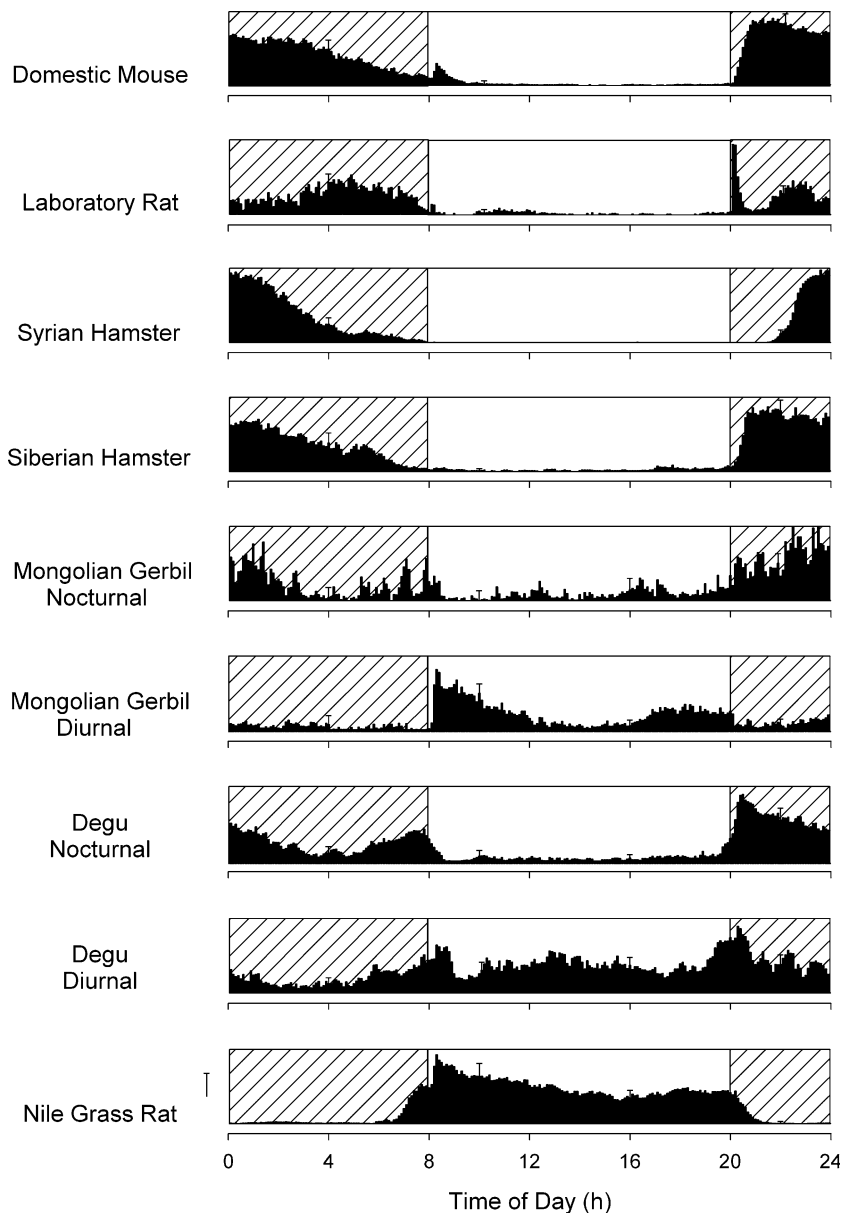
Fig. 3 Actogram of the running-wheel activity rhythm of a degu that spontaneously switched from a nocturnal to a diurnal pattern. Time of day is indicated on the horizontal axis and number of days on the vertical axis. The white and black bars above the actograms indicate the duration of the light and dark phases of the light–dark cycle, respectively

each species. Because of the segregation of Mongolian gerbils and degus into two groups, separate plots were prepared for animals classified as nocturnal and animals classified as diurnal in these two species. Inspection of the figure indicates that domestic mice, laboratory rats, Syrian hamsters, and Siberian hamsters are almost exclusively nocturnal. Nonetheless, some inter-species differences can be noted. Under LD 12:12, mice and Siberian hamsters initiate activity shortly after lights-off and run through most of the night. Laboratory rats are active immediately upon lights-off but run more erratically during the rest of the night. Syrian hamsters initiate activity about 2 h after lights-off and run for only 6–8 h each night. Nocturnal Mongolian gerbils are more active during the night than during the day but exhibit a considerable amount of activity during the light hours,

whereas diurnal gerbils initiate activity shortly after lights-on and exhibit little activity during the dark hours. Nocturnal degus exhibit an activity pattern similar to that of domestic mice, whereas the activity of diurnal degus is spread out through the day with peaks at dawn and dusk and greater activity during the light phase than during the dark phase. Finally, Nile grass rats are active throughout the light phase, initiating activity about an hour before lights-on and terminating activity about an hour after lights-off.

Not evident in Fig. 4 is the fact that Mongolian gerbils, degus, and Nile grass rats often exhibited bimodal behavioral patterns with increased activity around lights-on (dawn) and lights-off (dusk). Closer observation of Fig. 4 reveals a peak of activity at lights-on in diurnal gerbils, peaks prior to lights-on and after

Fig. 4 Average wave-form of the activity rhythms of the seven species, with separate plots for diurnal and nocturnal Mongolian gerbils and diurnal and nocturnal degus. Wave-forms were constructed by averaging 10 consecutive days for each animal and then averaging all animals in each species. Indices of inter-individual variability (standard errors of the means) are plotted only at 04:00, 10:00, 16:00, and 22:00 to prevent cluttering of the figure. *Hatching* indicates the dark phase of the light–dark cycle



lights-off in nocturnal degus, peaks after lights-on and at lights-off in diurnal degus, and a small peak at lights-on in grass rats. Because some individuals of each these species exhibited dawn, dusk, or dawn and dusk peaks of activity whereas others did not, the wave-form plots for the species are not as bimodal as the plots for some of the individuals.

Figure 5 shows frequency distributions of the scores of diurnality, onset time, and acrophase for the seven species. The three variables are generally consistent in each species. For instance, in domestic mice, the diurnality scores are clustered at the beginning of the dark phase, as are the onset times. As expected, the

acrophases lag behind the onsets by several hours. In Mongolian gerbils and degus, the distributions are rather spread out, essentially because of the segregation of individuals into diurnal and nocturnal groups.

The mean values of diurnality, onset time, acrophase, activity level, and robustness for the seven species are shown in Table 2. Analyses of variance were conducted separately for each of the five parameters. In all cases, a significant effect of species was identified ($F_{6, 183} > 40$, $P < 0.0001$). The significance of pairwise comparisons of means ($P < 0.05$ familywise) is indicated by the lowercase letters adjacent to the means. Lowercase letters are also used to denote significant differences in variance (square

Fig. 5 Frequency distributions of the scores of diurnality, onset time, and acrophase for the seven species. Onset time and acrophase are given in time of day (hours). *Hatching* indicates the dark phase of the light–dark cycle

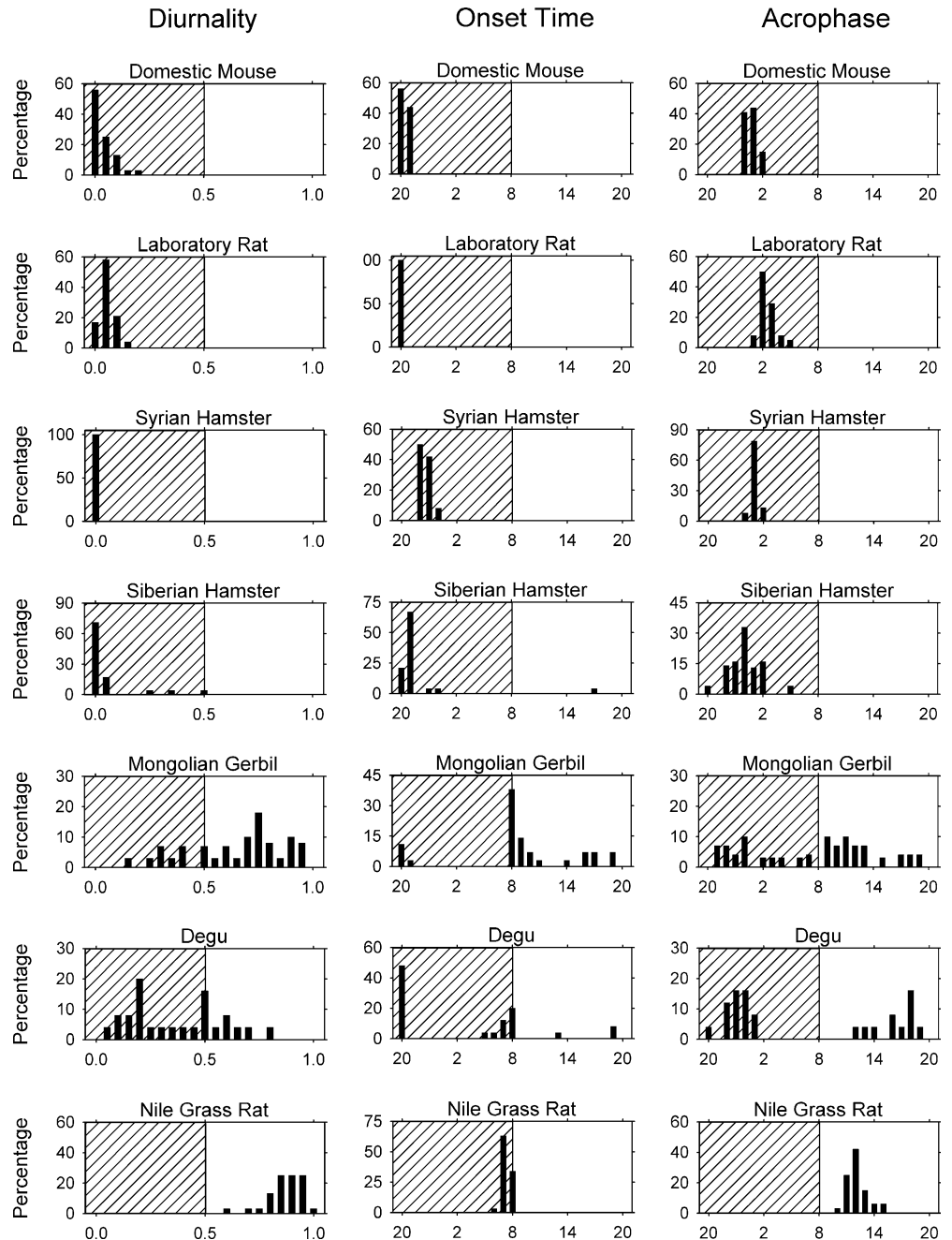


Table 2 Mean values of diurnality, onset time, acrophase, activity level, and robustness for the seven species

	Diurnality		Onset time (hours)		Acrophase (hours)		Activity (revolutions/day)		Robustness (Q_p at 24.0 h)	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Domestic mouse	0.039 a	0.049 a	20.5 a	0.3 a	24.7 a	0.6 a	14276 a	4483 a	1249 a	231 a
Laboratory rat	0.057 a	0.032 a	20.1 a	0.1 b	26.3 a	0.9 a	466 b	247 b	505 b	87 b
Syrian hamster	0.001 a	0.005 b	22.5 b	0.6 a	24.9 a	0.4 a	8267 c	2946 c	1669 c	277 a
Siberian hamster	0.057 a	0.127 c	20.8 a	1.6 c	24.1 a	1.6 b	5784 c	3847 a	1041 d	434 c
Mongolian gerbil	0.637 b	0.225 d	12.2 c	4.9 d	17.0 b	7.4 c	2002 d	1297 d	548 b	180 a
Nile grass rat	0.871 c	0.077 c	7.3 d	0.5 a	13.0 c	1.3 a	10253 e	6385 a	1027 d	361 c
Degu	0.363 d	0.228 d	14.4 c	6.3 d	20.1 d	4.0 c	9975 e	4805 a	733 b	307 c

For each parameter, means and standard deviations (SD) of species bearing the same letters (a, b, c, etc.) are not significantly different from each other ($P > 0.05$ familywise). Acrophase times are indicated in excess of 24.0 h where needed to allow statistical comparison with onset times

of standard deviation). Inspection of the table indicates that the first four species have mean diurnality scores much lower than 0.5 (and, therefore, are nocturnal). The mean diurnality index of 0.001 in Syrian hamsters indicates that only 0.1% of wheel revolutions occurred during the light phase of the LD cycle (or 99.9% occurred during the dark phase). Nile grass rats have a mean score much higher than 0.5 (and, therefore, are diurnal), whereas the mean score of 0.637 for Mongolian gerbils defines the species as predominantly diurnal, and the mean score of 0.363 for degus defines the species as predominantly nocturnal. The mean values of onset time and acrophase are generally consistent with the mean values of the diurnality index.

Table 2 also shows significant inter-species differences in the absolute levels of activity (wheel revolutions per day), the laboratory mouse being the most active species and the laboratory rat being the least active species. The comparison of variances for activity is somewhat misleading because the group means differ by more than an order of magnitude, and small means have proportionally smaller variances. A more meaningful comparison is based on the standard deviation as a

percentage of the mean. According to this criterion, inter-individual variability of activity is smaller in domestic mice (31%) and Syrian hamsters (36%) than in laboratory rats (53%), Siberian hamsters (66%), Mongolian gerbils (65%), Nile grass rats (62%), and degus (48%).

Although the laboratory mouse exhibited the highest activity level of all species, the activity rhythm of Syrian hamsters was more robust than that of mice (Table 2). The laboratory rat, which exhibited the lowest level of activity, exhibited also the least robust activity rhythm of all species.

The species means shown in Table 2 provide an accurate description of the rhythmic parameters in the seven species studied. However, given the bimodal distribution of diurnality in Mongolian gerbils and degus, a more meaningful inter-species comparison requires separate computations for the nine groups of animals. Figure 6 shows the mean diurnality scores for the nine groups. Nile grass rats are clearly diurnal, whereas laboratory rats, Siberian hamsters, domestic mice, and Syrian hamsters are clearly nocturnal. A downward gradient of diurnality is observed from Mongolian

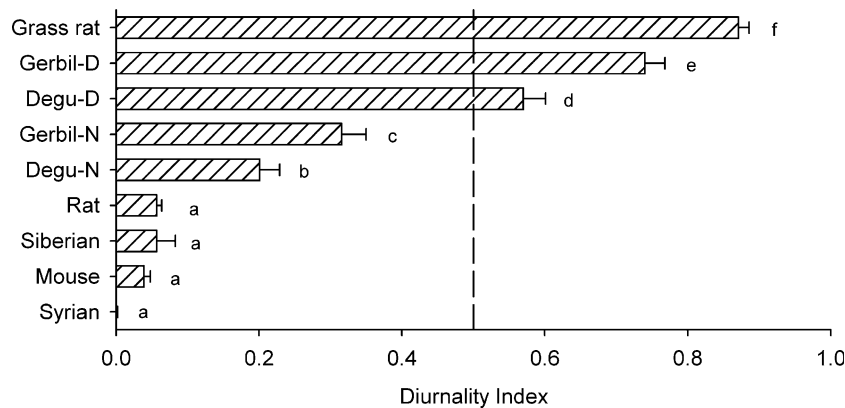


Fig. 6 Mean (\pm SE) diurnality of the nine groups of animals. Larger numbers reflect greater diurnality. Means of species bearing different letters (a, b, c, etc.) are significantly different from each other ($P < 0.05$ familywise). The dashed line indicates the theoretical

separation between nocturnal and diurnal animals (i.e., equal amounts of activity during the light and dark phases of the light-dark cycle)

Table 3 Correlation coefficients

	Activity vs. robustness	Diurnality vs. onset	Diurnality vs. acrophase	Onset vs. acrophase
Domestic mouse	+0.66*	+0.25	+0.64*	+0.26
Laboratory rat	+0.30	-0.12	+0.50	-0.28
Syrian hamster	+0.54	-0.20	-0.06	+0.37
Siberian hamster	+0.73*	-0.49	-0.41	+0.84*
Mongolian gerbil	+0.50	-0.69*	-0.41	+0.18
Nile grass rat	+0.84*	+0.39	-0.37	+0.18
Degu	+0.41	-0.83*	-0.84*	+0.69*

* $P < 0.001$ (for overall $P < 0.05$)

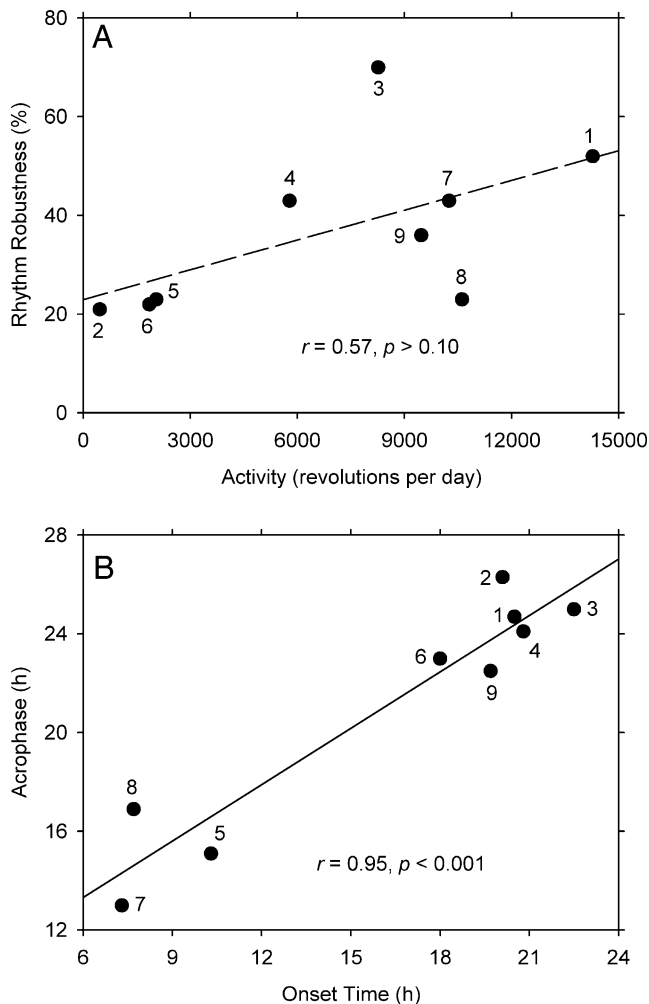


Fig. 7 Inter-species correlations for amount of activity and rhythm robustness (a) and for onset time and acrophase time (b). Robustness is expressed as percentage of maximum Q_p (with percentages greater than 13% being significantly above noise level). Acrophase times are indicated in excess of 24.0 h where needed to allow comparison with onset times. The numbers adjacent to the data points identify the various species, as follows: 1 domestic mouse, 2 laboratory rat, 3 Syrian hamster, 4 Siberian hamster, 5 Mongolian gerbil (diurnal), 6 Mongolian gerbil (nocturnal), 7 Nile grass rat, 8 Degu (diurnal), and 9 Degu (nocturnal)

gerbils classified as diurnal, degus classified as diurnal, gerbils classified as nocturnal, and degus classified as nocturnal.

Intra-species correlation coefficients were computed for activity level (revolutions per day), diurnality index, onset time, acrophase, and rhythm robustness, as shown in Table 3. Although some significant correlations were found for some pairs of parameters in some species, no general pattern emerged. In particular, a high correlation between onset time and acrophase would have been expected. However, because the intra-species ranges of variability were quite narrow in all parameters, the lack of correlation can be considered to be a statistical artifact. More meaningful comparisons can be made across species. Figure 7 shows two inter-species correlation plots. Rhythm robustness did not correlate significantly with activity level across species (as it did not within species), but acrophase and onset time were significantly correlated across species ($r = 0.95, P < 0.001$). That is, the activity rhythms of species that initiated activity later in the day had acrophases later in the day, as expected.

Because the robustness of the rhythm of body temperature in laboratory rodents seems to be greater in larger species (Refinetti 1996a), a coefficient correlation for body mass and robustness of the running-wheel activity rhythm was computed. The coefficient was found to be negative ($r = -0.42$) and not statistically significant ($P = 0.355$).

To verify the appropriateness of the use of running wheels for the monitoring of locomotor activity, selected animals were tested with an infrared motion detector. Six degus that were clearly nocturnal during the regular study with running wheels were transferred to cages with infrared motion detectors. Activity was monitored while the wheels remained available and after the wheels were removed. In none of the six cases was there a reversal of the activity pattern. As shown in Fig. 8, removal of the wheel had no apparent effect on the activity pattern of the animals. Also, both nocturnal and diurnal degus and Mongolian gerbils were placed in constant darkness for several weeks at the end of the study (with activity monitored by running wheels). No abrupt change in the phase of the activity rhythm (indicative of a masking effect of the previous LD cycle) was observed in either species. Degus and Mongolian gerbils free-ran with mean (\pm SE) circadian periods of 23.73 ± 0.07 h and 24.24 ± 0.04 h, respectively.

To further investigate the differences between diurnal and nocturnal degus, 12 individuals (7 diurnal,

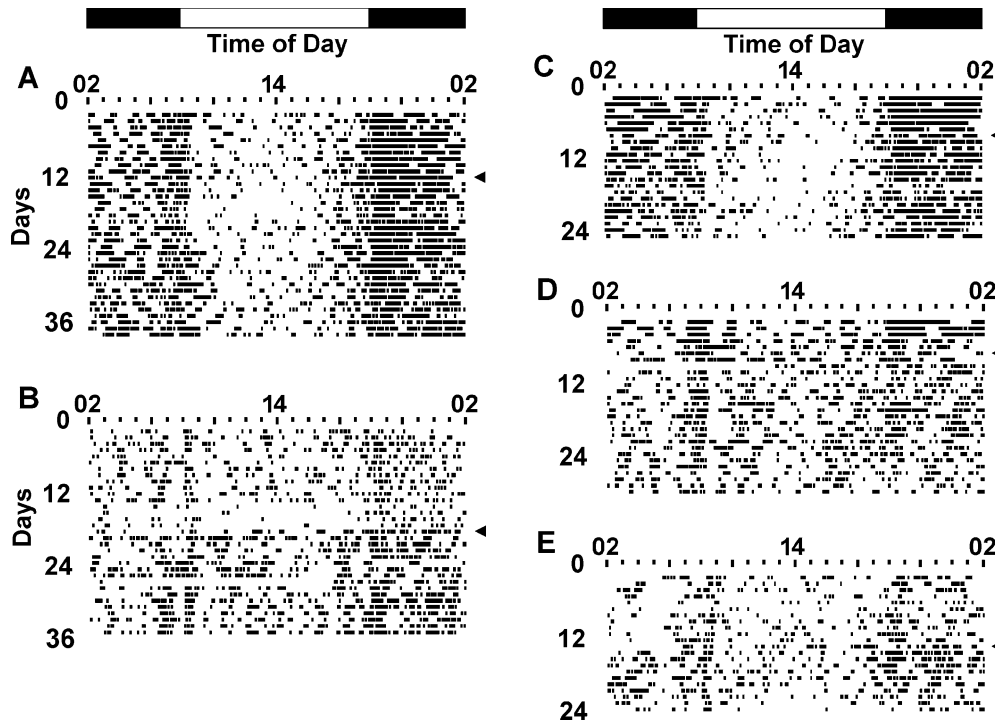


Fig. 8 Records of locomotor activity of five degus obtained by an infrared motion detector while the animals had access to a running wheel and after the wheel was removed (as indicated by the arrowheads on the right margin). Time of day is indicated on the horizontal axis and number of days on the vertical axis. Only bins with five or more breaks of the infrared beam are plotted. Because

activity levels were consistently lower without the wheel, the actograms are plotted in digital rather than analog format (that is, for each 6-min bin, a pen mark appears if there were five or more breaks of the infrared beam; otherwise, the space is left blank). The white and black bars above the actograms indicate the duration of the light and dark phases of the light–dark cycle, respectively

5 nocturnal) were transferred to an ultradian LD cycle (LD 3.5:3.5) for the evaluation of photic masking responses. Figure 9 shows records of two representative animals. Visual inspection of records obtained under short LD cycles is very difficult, but it appears that the diurnal degu exhibited slightly greater activity during the light phase of the LD 3.5:3.5 cycle than did the nocturnal degu. For quantitative analysis, data from the first 3 days under LD 3.5:3.5 were discarded and data from the following 10 days were used for computation of the diurnality index, as described above ($R_L/[R_L + R_D]$). The seven diurnal degus exhibited a mean diurnality index (\pm SE) of 0.59 ± 0.07 under LD 3.5:3.5, whereas the five nocturnal degus exhibited a mean diurnality index of 0.32 ± 0.05 . The two means are significantly different from each other ($t_{10} = 2.696$, $P = 0.022$). Although the mean index for diurnal degus is not different from that exhibited under LD 12:12 ($t_{16} = 0.231$, $P > 0.80$), the mean index for nocturnal degus is marginally larger than that exhibited under LD 12:12 ($t_{17} = 2.155$, $P = 0.044$), which indicates a lesser degree of nocturnality.

For comparative purposes, four Syrian hamsters were also transferred to LD 3.5:3.5. The mean diurnality index for these hamsters was 0.13 ± 0.01 , which is significantly larger than the mean index under LD 12:12 [0.001 ± 0.0003 ($t_{26} = 24.137$, $P < 0.001$)].

Discussion

Domestic mice, laboratory rats, Syrian hamsters, and Siberian hamsters are traditionally considered nocturnal species, and the results of this study are consistent with this view. In these four species, 94% or more of daily activity (wheel revolutions) occurred during the dark phase of the LD cycle, and the daily onset of activity took place at or shortly after the time of lights-off. This predominantly nocturnal pattern of activity is consistent with hundreds of previous observations in domestic mice (e.g., Davis and Menaker 1981; Kramer et al. 1998; Refinetti 2004a; Weinert and Waterhouse 1998), laboratory rats (e.g., Francis and Coleman 1988; Honma and Hiroshige 1978; Ikeda and Inoué 1998; Meinrath and D'Amato 1979), Syrian hamsters (e.g., Boulou et al. 1996; Conn et al. 1990; DeCoursey et al. 1998; Weinert et al. 2001), and Siberian hamsters (e.g., Anchordoquy and Lynch 2000; Ruby et al. 2002 b; Steinlechner et al. 2002; Tokura and Oishi 1985). Nonetheless, this study evinced both intra- and inter-species differences in the strength of nocturnality. On average for these four species, the intra-species standard deviation of onset times was 38 min and the standard deviation of acrophases was 53 min. Inter-species differences in diurnality among these four species were small and not statistically

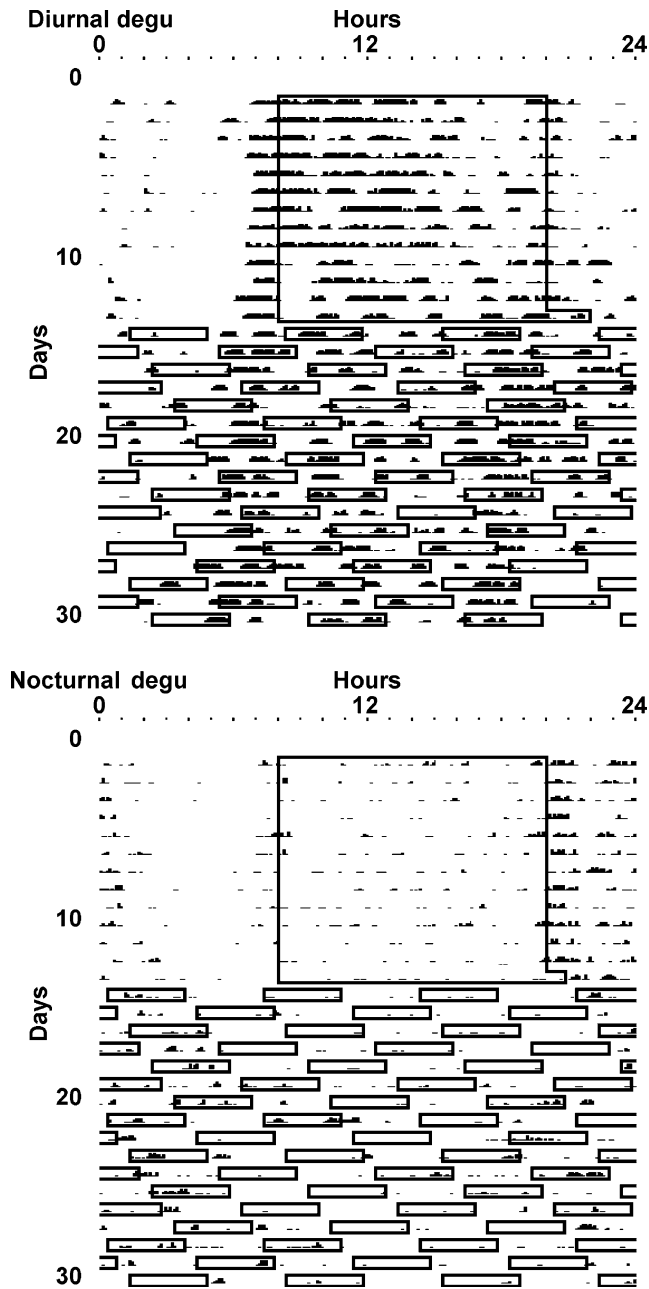


Fig. 9 Actograms of the running-wheel activity rhythms of a diurnal and a nocturnal degu maintained first under LD 12:12 and later under LD 3.5:3.5. Time of day is indicated on the horizontal axis and number of days on the vertical axis. The rectangles indicate the intervals of lights-on

significant, although 99.9% of the running-wheel activity of Syrian hamsters was restricted to the dark phase, whereas only 94.3% of the running-wheel activity of laboratory rats and Siberian hamsters was restricted to the dark phase. The robustness of the activity rhythm was significantly lower in laboratory rats (21% of maximum) than in domestic mice (52%), Syrian hamsters (70%), and Siberian hamsters (43%), although this was not accompanied by greater intra-species variability in onset times, acrophase, or diurnality index.

Mongolian gerbils showed great intra-species variability, with 24% of the animals being predominantly nocturnal and 76% being predominantly diurnal. Within the two subgroups of gerbils, rhythm robustness was lower—and the variabilities in onset time, acrophase, and diurnality were greater—than in mice, rats, and hamsters. Circadian rhythms of Mongolian gerbils had not been previously studied in detail. A study limited to two specimens, in which activity was monitored by contact sensors in a cage without running wheels, found that Mongolian gerbils were predominantly crepuscular with a tendency to nocturnality (Nelissen and Nelissen-Joris 1975). In a study on a single gerbil, in which activity was also monitored by contact sensors, the activity pattern was very erratic with a weak crepuscular component (Probst 1992). In a study on four gerbils in which activity was monitored by radio-telemetry, the acrophase of the activity rhythm was found to lie midway between the mean acrophases of nocturnal and diurnal species (Refinetti 1999). Thus, although the current finding of a greater proportion of diurnal than of nocturnal gerbils in a sample of 29 individuals is novel, it does not contradict previous observations. Because the infrared motion detectors were not available at the time when the gerbils were tested, it is not possible to ascertain whether the large proportion of diurnal gerbils obtained here is associated with the use of running wheels for the monitoring of activity.

Degus were similar to gerbils in that the robustness of their activity rhythms (31% of maximum) was significantly lower than that of mice and hamsters and in that some individuals were diurnal whereas others were nocturnal. Although degus are considered to be diurnal in the wild (Kenagy et al. 2002) and in the laboratory if monitored by radio-telemetry (Refinetti 1996b), large inter-individual variability in the phase angle of entrainment has been reported in degus monitored by radio-telemetry (Labyak et al. 1997). In one laboratory, it was found that some degus were diurnal without running wheels but became nocturnal when given access to wheels (Kas and Edgar 1999). The degus in the present study showed both diurnal and nocturnal patterns of activity when running wheels were available, but the patterns were not reversed when activity was recorded with infrared motion detectors. Thus, diurnal and nocturnal patterns of activity were associated with different individuals, not with different recording methods. The explanation for the discrepancy between the two studies is not evident. One possible explanation for the absence of phase reversals when the wheel was removed in the present study is that the animals were not studied for a sufficiently long interval of time. This is very unlikely, however, because some animals were studied for up to 3 weeks after the removal of the wheels, and Kas and Edgar reported reversals on the day immediately following the transition. The present observations that degus may be diurnal or nocturnal regardless of recording methods and that some degus spontaneously switch niches might explain a few cases of

niche switching in Kas and Edgar's study as mere coincidences unrelated to the removal or presentation of running wheels. However, Kas and Edgar reported observing the phenomenon in seven distinct individuals, which makes an explanation based on coincidence very unlikely. Whatever the true explanation may be, it is important to point out that the discrepancy is limited to the phenomenon of wheel-induced niche switches. The two studies are in agreement about the observation that approximately half of the degus are naturally diurnal (44% in this study, 54% in Kas and Edgar's) and half are naturally nocturnal (56 and 46%, respectively).

Nile grass rats exhibited robust activity rhythms with a predominantly diurnal pattern. On average, 87% of daily activity (wheel revolutions) occurred during the light phase of the LD cycle, and the daily onset of activity took place approximately 1 h before the time of lights-on. Appearance of nocturnal activity patterns in animals given access to running wheels has been reported in Nile grass rats (Blanchong et al. 1999), but it is likely that the animals that were observed to be nocturnal when given access to running wheels were members of an idiosyncratic subgroup. In my laboratory, all grass rats housed with running wheels have been diurnal (Refinetti 2004a, c). A third research team observed increased crepuscular activity in animals housed with wheels, but the activity pattern was still predominantly diurnal (Redlin and Mrosovsky 2004). Observations in the wild in Kenya were consistent with the data from these laboratories in revealing a predominantly diurnal pattern of activity (Blanchong and Smale 2000).

In summary, domestic mice, laboratory rats, Syrian hamsters, and Siberian hamsters were found to be predominantly nocturnal, with small intra- and inter-species variability. Nile grass rats were found to be predominantly diurnal, again with small intra-species variability. Degus and Mongolian gerbils were found to be either diurnal or nocturnal. Great intra-species variability in diurnality, with some individuals showing predominantly diurnal activity patterns and others showing predominantly nocturnal activity patterns, has been occasionally described in other species. For instance, in goldfish (*Carassius auratus*), about 80% of individuals tested in the laboratory were found to be diurnal, whereas 10% were nocturnal, and 10% displayed very weak rhythmicity (Iigo and Tabata 1996). In carpenter ants (*Camponotus compressus*), approximately 70% of individually tested animals were found to be nocturnal, whereas 30% were diurnal (Sharma et al. 2004). Likewise, in subterranean mole rats of various species, some members of the species were found to be diurnal and some were found to be nocturnal (Oosthuizen et al. 2003; Oster et al. 2002). Even some instances of intra-individual variability (i.e., the same individual being diurnal under some circumstances and nocturnal under other circumstances) have been previously reported. For instance, wolves (*Canis lupus*) are normally nocturnal; however, when traveling over long distances, they travel during the day (Merrill and Mech 2003). Conversely,

migratory birds are normally diurnal, but they do most of their migratory flight at night (Cochran 1987; Rattenborg et al. 2004). The present results do not confirm the previous report of wheel-induced reversal of diurnality in degus (Kas and Edgar 1999).

Intra-species and intra-individual variabilities in diurnality complicate further the distinction between diurnal and nocturnal species. Clearly, the diurnal–nocturnal dichotomy reflects researchers' needs for systematization more than a consistent distinction between species. It has been proposed that the distinction between diurnality and nocturnality depends on two relatively independent processes: the establishment of an adequate phase relationship between the circadian clock and activity (that is, proper entrainment) and the establishment of an adequate pattern of photic masking (that is, negative masking in nocturnal organisms and positive masking in diurnal organisms) (Mrosovsky 1999). Regarding the phase relationship between the circadian clock and activity, studies on mutant mice and hamsters with abnormally long or short endogenous periods have shown that animals with a typically nocturnal circadian system will entrain to the LD cycle with a diurnal phase angle if the characteristics of the LD cycle so require (Low-Zeddies and Takahashi 2001; Ralph and Menaker 1988). That is, when the circadian period is lengthened or shortened without a concomitant change in the photic phase–response curve, entrainment to a standard LD cycle can only be attained by longer exposure to light, which requires a shift of the active phase into daylight. Regarding masking, numerous studies have shown that light exposure during the night often inhibits activity in nocturnal animals and elicits activity in diurnal animals (Aschoff and von Goetz 1989; Edelstein and Mrosovsky 2001; Mrosovsky et al. 1999; Mrosovsky and Hattar 2003; Redlin and Mrosovsky 1999, 2004). In a recent review article, Mrosovsky and Hattar (2005) described several studies in which the temporal niche of mice was switched from nocturnal to diurnal as a result of genetic manipulations of photic receptor systems (rod and cone opsins, cryptochrome, and melanopsin). Mice deficient in photic receptors exhibited much more activity during the light phase than did normal mice and also often exhibited positive masking in response to short pulses of light. The appearance of positive masking in a species that usually exhibits negative masking suggests that the niche inversion was achieved at least partially by an inversion in masking mechanisms.

If the distinction between diurnality and nocturnality indeed depends on the interplay of mechanisms of entrainment and masking, then it is possible that intermediate forms of diurnality (as observed in Mongolian gerbils and degus in the present study) result from unusual combinations of patterns of entrainment and masking. The data from degus exposed to ultradian cycles (LD 3.5:3.5) provided modest support for this hypothesis. Because LD 3.5:3.5 provides photic masking but not entrainment, whereas LD 12:12 provides both

masking and entrainment, the equivalency of diurnality indexes in diurnal degus (0.59 and 0.57, respectively) suggests that diurnality in this species is determined mostly by masking. In nocturnal degus, the diurnality index was not as small under LD 3.5:3.5 (0.32) as under LD 12:12 (0.20), which suggests that entrainment plays a relatively greater role in the establishment of nocturnality. Nonetheless, the fact that the diurnality index under LD 3.5:3.5 was greater than 0.5 in diurnal degus and smaller than 0.5 in nocturnal degus is consistent with the notion of positive masking in diurnal animals and negative masking in nocturnal animals. The present results provide no indication of a gradual (incomplete) inversion of masking mechanisms in degus.

In an attempt to understand the mechanisms responsible for the choice of temporal niche, several recent studies compared the neural circuits involved in the control of circadian rhythms in the predominantly diurnal Nile grass rat and the predominantly nocturnal laboratory rat. The operation of the circadian pacemaker, as assessed by measures of gene expression in the brain, was found to be essentially identical in the two species, and any observed differences were inferred to take place downstream from the pacemaker (Mahoney et al. 2001; Novak et al. 2000; Nunez et al. 1999; Schwartz et al. 2004; Lambert et al. 2005). Furthermore, in guinea pigs (*Cavia porcellus*), which exhibit an activity pattern that is not clearly diurnal or nocturnal, electrophysiological recording of brain cells indicates robust rhythmicity in the circadian pacemaker (with greater activity during the day) despite the poorly differentiated pattern of behavioral activity (Kurumiya and Kawamura 1988). Two recent studies from one laboratory suggested that there may be differences between diurnal and nocturnal rodents in the operation of the pacemaker during subjective day (Novak and Albers 2004a, b). However, because the differences were small and were identified in the comparison of only two species (the Nile grass rat and the golden hamster), further studies are necessary before any generalizations can be made.

In conclusion, the diurnal–nocturnal dichotomy is inadequate to describe the variety of circadian chronotypes exhibited by different species. Even in the small sample of seven rodent species used in this study, a wide range of diurnal and nocturnal patterns of activity was observed. Through mechanisms as yet poorly understood, the balance between entraining and masking processes seems to generate a gradient of temporal niches that runs from predominantly diurnal species to predominantly nocturnal species with many chronotypes in between, including species that exhibit wide intraspecies gradients of temporal niche.

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References

- Anchordoquy HC, Lynch GR (2000) Timing of testicular recrudescence in Siberian hamsters is unaffected by pinealectomy or long-day photoperiod after 9 weeks in short days. *J Biol Rhythms* 15:406–416
- Aschoff J (1966) Circadian activity pattern with two peaks. *Ecology* 47:657–662
- Aschoff J, von Goetz C (1989) Masking of circadian activity rhythms in canaries by light and dark. *J Biol Rhythms* 4:29–38
- Blanchong JA, Smale L (2000) Temporal patterns of activity of the unstriped Nile rat, *Arvicanthis niloticus*. *J Mammal* 81:595–599
- Blanchong JA, McElhinny TL, Mahoney MM, Smale L (1999) Nocturnal and diurnal rhythms in the unstriped Nile rat, *Arvicanthis niloticus*. *J Biol Rhythms* 14:364–377
- Boulos Z, Macchi M, Houpt TA, Terman M (1996) Photic entrainment in hamsters: effects of simulated twilights and nest box availability. *J Biol Rhythms* 11:216–233
- Cochran WW (1987) Orientation and other migratory behaviours of a Swainson's thrush followed for 1500 km. *Anim Behav* 35:927–928
- Conn CA, Borer KT, Kluger MJ (1990) Body temperature rhythm and response to pyrogen in exercising and sedentary hamsters. *Med Sci Sports Exerc* 22:636–642
- Dardente H, Menet JS, Challet E, Tournier BB, Pévet P, Masson-Pévet M (2004) Daily and circadian expression of neuropeptides in the suprachiasmatic nuclei of nocturnal and diurnal rodents. *Mol Brain Res* 124:143–151
- Davis FC, Menaker M (1981) Development of the mouse circadian pacemaker: independence from environmental cycles. *J Comp Physiol A* 143:527–539
- DeCoursey PJ, Pius S, Sandlin C, Wethey D, Schull J (1998) Relationship of circadian temperature and activity rhythms in two rodent species. *Physiol Behav* 65:457–463
- Edelstein K, Mrosovsky N (2001) Behavioral responses to light in mice with dorsal lateral geniculate lesions. *Brain Res* 918:107–112
- Engelmann W (1988) Evolution and selective advantage of circadian rhythms. *Acta Physiol Pol* 39:345–356
- Fidler AE, Gwinner E (2003) Comparative analysis of avian BMAL1 and CLOCK protein sequences: a search for features associated with owl nocturnal behavior. *Comp Biochem Physiol B* 136:861–874
- Foster RG, Provencio I, Hudson D, Fiske S, De Grip W, Menaker M (1991) Circadian photoreception in the retinally degenerate mouse (rd/rd). *J Comp Physiol A* 169:39–50
- Francis AJP, Coleman GJ (1988) The effect of ambient temperature cycles upon circadian running and drinking activity in male and female laboratory rats. *Physiol Behav* 43:471–477
- Freedman MS, Lucas RJ, Soni B, von Schantz M, Muñoz M, David-Gray Z, Foster R (1999) Regulation of mammalian circadian behavior by non-rod, non-cone, ocular photoreceptors. *Science* 284:502–504
- Goldman BD, Goldman SL, Riccio AP, Terkel J (1997) Circadian patterns of locomotor activity and body temperature in blind mole-rats, *Spalax ehrenbergi*. *J Biol Rhythms* 12:348–361
- Gooley JJ, Lu J, Fischer D, Saper CB (2003) A broad role for melanopsin in nonvisual photoreception. *J Neurosci* 23:7093–7106
- Halberg F (1953) Some physiological and clinical aspects of 24-hour periodicity. *J Lancet* 73:20–32
- Hattar S, Liao HW, Takao M, Berson DM, Yau KW (2002) Melanopsin-containing retinal ganglion cells: architecture, projections, and intrinsic photosensitivity. *Science* 295:1065–1069
- Hays WL (1988) *Statistics*, 4th edn. Holt, Rinehart & Winston, New York
- Hofstetter JR, Hofstetter AR, Hughes AM, Mayeda AR (2005) Intermittent long-wavelength red light increases the period of daily locomotor activity in mice. *J Circadian Rhythms* 3:8

- Honma K, Hiroshige T (1978) Simultaneous determination of circadian rhythms of locomotor activity and body temperature in the rat. *Jpn J Physiol* 28:159–169
- Iigo M, Tabata M (1996) Circadian rhythms of locomotor activity in the goldfish *Carassius auratus*. *Physiol Behav* 60:775–781
- Ikeda M, Inoué S (1998) Simultaneous recording of circadian rhythms of brain and intraperitoneal temperatures and locomotor and drinking activities in the rat. *Biol Rhythm Res* 29:142–150
- Kas MJH, Edgar DM (1999) A nonphotic stimulus inverts the diurnal–nocturnal phase preference in *Octodon degus*. *J Neurosci* 19:328–333
- Kenagy GJ, Nespolo RF, Vásquez RA, Bozinovic F (2002) Daily and seasonal limits of time and temperature to activity of degus. *Rev Chil Hist Nat* 75:567–581
- Kirk RE (1995) Experimental design: procedures for the behavioral sciences, 3rd edn. Brooks/Cole, Pacific Grove
- Klerman EB, Shanahan TL, Brotman DJ, Rimmer DW, Emens JS, Rizzo JF, Czeisler CA (2002) Photic resetting of the human circadian pacemaker in the absence of conscious vision. *J Biol Rhythms* 17:548–555
- Kramer K, Voss HP, Grimbergen J, Bast A (1998) Circadian rhythms of heart rate, body temperature, and locomotor activity in freely moving mice measured with radio telemetry. *Lab Anim* 27(8):23–26
- Kurumiya S, Kawamura H (1988) Circadian oscillation of the multiple unit activity in the guinea pig suprachiasmatic nucleus. *J Comp Physiol A* 162:301–308
- Labyak SE, Lee TM, Goel N (1997) Rhythm chronotypes in a diurnal rodent, *Octodon degus*. *Am J Physiol* 273:R1058–R1066
- Lambert CM, Machida KK, Smale L, Nunez AA, Weaver DR (2005) Analysis of the prokineticin 2 system in a diurnal rodent, the unstriped Nile grass rat (*Arvicanthis niloticus*). *J Biol Rhythms* 20:206–218
- Lincoln G, Messenger S, Andersson H, Hazlerigg D (2002) Temporal expression of seven clock genes in the suprachiasmatic nucleus and the pars tuberalis of the sheep: evidence for an internal coincidence timer. *Proc Natl Acad Sci USA* 99:13890–13895
- Low-Zeddies SS, Takahashi JS (2001) Chimera analysis of the Clock mutation in mice shows that complex cellular integration determines circadian behavior. *Cell* 105:25–42
- Mahoney M, Bult A, Smale L (2001) Phase response curve and light-induced Fos expression in the suprachiasmatic nucleus and adjacent hypothalamus of *Arvicanthis niloticus*. *J Biol Rhythms* 16:149–162
- Marques MD, Waterhouse JM (1994) Masking and the evolution of circadian rhythmicity. *Chronobiol Int* 11:146–155
- Meinrath M, D'Amato MR (1979) Interrelationships among heart rate, activity, and body temperature in the rat. *Physiol Behav* 22:491–498
- Merrill SB, Mech LD (2003) The usefulness of GPS telemetry to study wolf circadian and social activity. *Wildl Soc Bull* 31:947–960
- Mrosovsky N (1999) Masking: history, definitions, and measurement. *Chronobiol Int* 16: 415–429
- Mrosovsky N (2003a) Aschoff's rule in retinally degenerate mice. *J Comp Physiol A* 189:75–78
- Mrosovsky N (2003b) Beyond the suprachiasmatic nucleus. *Chronobiol Int* 20:1–8
- Mrosovsky N, Hattar S (2003) Impaired masking responses to light in melanopsin-knockout mice. *Chronobiol Int* 20:989–999
- Mrosovsky N, Hattar S (2005) Diurnal mice (*Mus musculus*) and other examples of temporal niche switching. *J Comp Physiol A* 191:1011–1024
- Mrosovsky N, Foster RG, Salmon PA (1999) Thresholds for masking responses to light in three strains of retinally degenerate mice. *J Comp Physiol A* 184:423–428
- Nelissen M, Nelissen-Joris N (1975) On the diurnal rhythm of activity of *Meriones unguiculatus* (Milne-Edwards, 1867). *Acta Zool Pathol Antwerp* 61:25–30
- Nelson W, Tong YL, Lee JK, Halberg F (1979) Methods for cosinor rhythmometry. *Chronobiologia* 6:305–323
- Novak CM, Albers HE (2004a) Novel phase-shifting effects of GABA_A receptor activation in the suprachiasmatic nucleus of a diurnal rodent. *Am J Physiol* 286:R820–R825
- Novak CM, Albers HE (2004b) Circadian phase alteration by GABA and light differs in diurnal and nocturnal rodents during the day. *Behav Neurosci* 118:498–504
- Novak CM, Harris JA, Smale L, Nunez AA (2000) Suprachiasmatic nucleus projections to the paraventricular thalamic nucleus in nocturnal rats (*Rattus norvegicus*) and diurnal Nile grass rats (*Arvicanthis niloticus*). *Brain Res* 874:147–157
- Nunez AA, Bult A, McElhinny TL, Smale L (1999) Daily rhythms of Fos expression in hypothalamic targets of the suprachiasmatic nucleus in diurnal and nocturnal rodents. *J Biol Rhythms* 14:300–306
- Oosthuizen MK, Cooper HM, Bennett NC (2003) Circadian rhythms of locomotor activity in solitary and social species of African mole-rats (family: Bathyergidae). *J Biol Rhythms* 18:481–490
- Oster H, Avivi A, Joel A, Albrecht U, Nevo E (2002) A switch from diurnal to nocturnal activity in *S. ehrenbergi* is accompanied by an uncoupling of light input and the circadian clock. *Curr Biol* 12:1919–1922
- Panda S, Sato TK, Castrucci AM, Rollag MD, DeGrip WJ, Hogenesch JB, Provencio I, Kay SA (2002). Melanopsin (Opn4) requirement for normal light-induced circadian phase shifting. *Science* 298:2213–2216
- Probst B (1992) An automated method for recording scent marking in Mongolian gerbils. *Physiol Behav* 52:661–663
- Ralph MR, Menaker M (1988) A mutation of the circadian system in golden hamsters. *Science* 241:1225–1227
- Rattenborg NC, Mandt BH, Obermeyer WH, Winsauer PJ, Huber R, Wikelski M, Benca RM (2004) Migratory sleeplessness in the white-crowned sparrow (*Zonotrichia leucophrys gambelii*). *PLOS Biol* 2:924–936
- Redlin U, Mrosovsky N (1999) Masking of locomotor activity in hamsters. *J Comp Physiol A* 184:429–437
- Redlin U, Mrosovsky N (2004) Nocturnal activity in a diurnal rodent (*Arvicanthis niloticus*): the importance of masking. *J Biol Rhythms* 19:58–67
- Refinetti R (1996a) Comparison of the body temperature rhythms of diurnal and nocturnal rodents. *J Exp Zool* 275:67–70
- Refinetti R (1996b) Rhythms of body temperature and temperature selection are out of phase in a diurnal rodent, *Octodon degus*. *Physiol Behav* 60:959–961
- Refinetti R (1999) Relationship between the daily rhythms of locomotor activity and body temperature in eight mammalian species. *Am J Physiol* 277:R1493–R1500
- Refinetti R (2004a) Daily activity patterns of a nocturnal and a diurnal rodent in a seminatural environment. *Physiol Behav* 82:285–294
- Refinetti R (2004b) Non-stationary time series and the robustness of circadian rhythms. *J Theor Biol* 227:571–581
- Refinetti R (2004c) Parameters of photic resetting of the circadian system of a diurnal rodent, the Nile grass rat. *Acta Sci Vet* 32:1–6
- Refinetti R (2004d) The Nile grass rat as a laboratory animal. *Lab Anim* 33(9):54–57
- Roenneberg T, Mrosovsky N (2002) Life before the clock: modeling circadian evolution. *J Biol Rhythms* 17:495–505
- Ruby NF, Brennan TJ, Xie X, Cao V, Franken P, Heller HC, O'Hara BF (2002a) Role of melanopsin in circadian responses to light. *Science* 298:2211–2213
- Ruby NF, Joshi N, Heller HC (2002b) Constant darkness restores entrainment to phase-delayed Siberian hamsters. *Am J Physiol* 283:R1314–R1320
- Rutter J, Reick M, McKnight SL (2002) Metabolism and the control of circadian rhythms. *Ann Rev Biochem* 71:307–331
- Schwartz MD, Nunez AA, Smale L (2004) Differences in the suprachiasmatic nucleus and lower subparaventricular zone of diurnal and nocturnal rodents. *Neuroscience* 127:13–23

- Semo M, Peirson S, Lupi D, Lucas RJ, Jeffery G, Foster RG (2003) Melanopsin retinal ganglion cells and the maintenance of circadian and pupillary responses to light in aged rodless/coneless (rd/rd cl) mice. *Eur J Neurosci* 17:1793–1801
- Sharma VK, Lone SR, Mathew D, Goel A, Chandrashekar MK (2004) Possible evidence for shift work schedules in the media workers of the ant species *Camponotus compressus*. *Chronobiol Int* 21:297–308
- Smale L, Lee T, Nunez AA (2003) Mammalian diurnality: some facts and gaps. *J Biol Rhythms* 18:356–366
- Sokolove PG, Bushell WN (1978) The chi square periodogram: its utility for analysis of circadian rhythms. *J Theor Biol* 72:131–160
- Steinlechner S, Stieglitz A, Ruf T (2002) Djungarian hamsters: a species with a labile circadian pacemaker? Arrhythmicity under a light–dark cycle induced by short light pulses. *J Biol Rhythms* 17:248–258
- Tokura H, Oishi T (1985) Circadian locomotor activity rhythm under the influences of temperature cycle in the Djungarian hamster, *Phodopus sungorus*, entrained by 12 hour light–12 hour dark cycle. *Comp Biochem Physiol A* 81:271–275
- Weinert D, Waterhouse J (1998) Diurnally changing effects of locomotor activity on body temperature in laboratory mice. *Physiol Behav* 63:837–843
- Weinert D, Fritzsche P, Gattermann R (2001) Activity rhythms of wild and laboratory golden hamsters (*Mesocricetus auratus*) under entrained and free-running conditions. *Chronobiol Int* 18:921–932
- Yoshimura T, Yokota Y, Ishikawa A, Yasuo S, Hayashi N, Suzuki T, Okabayashi N, Namikawa T, Ebihara S (2002) Mapping quantitative trait loci affecting circadian photosensitivity in retinally degenerate mice. *J Biol Rhythms* 17:512–519