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Diel and circadian rhythms of locomotor activity in male *Parasteatoda tepidariorum* (Araneae: Theridiidae)

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Abstract. Despite recent interest, there still is relatively little known about the ecology and physiology of diel and circadian rhythms in spiders. However, previous work on spiders suggests that there is a striking amount of variation in circadian period both among, and within, species, when compared to model organisms. Whereas previous studies of behavioral rhythms in spiders focused on females, here we describe the diel and circadian patterns of locomotor activity in male *Parasteatoda tepidariorum* (C. L. Koch, 1841) (Theridiidae). We found that the males showed mostly nocturnal activity under a light:dark cycle, with activity peaking very early after lights off and steadily declining to near zero just prior to lights on. Under constant darkness most individuals showed significant circadian rhythmicity with a mean free-running period of about 21.2 h. Though not the shortest average free-running period described for spiders, being so out of resonance with the 24 h solar day strains conventional circadian rhythm theory. Our data also suggest that the phase angle of entrainment for locomotor activity is in the mid-to-late photophase, but that activity may be masked by light. Of particular note is that both the diel and circadian activity patterns reported here for male *P. tepidariorum* are similar to those reported elsewhere for females of the species. This study deepens our understanding of the nature and variation in circadian rhythm in spiders and builds a case for further developing spiders as a model system for research integrating the fields of chronobiology and ecology.

Keywords: Behavioral rhythm, circadian clock, chronoecology, circadian masking, wild clocks

Circadian rhythms are generated by endogenous clocks with periods close to 24 h. These clocks are temperature compensated, entrain to (synchronize with) the solar day, yet persist without an external time cue (Pittendrigh 1960), and are generally ubiquitous among animals (Johnson & Kondo 2001). The circadian clock influences most aspects of an organism's physiology and behavior (reviewed in Panda et al. 2002). In spiders specifically, the circadian clock has been shown to influence aspects of behavior and physiology such as locomotor activity (Seyfarth 1980; Suter 1993; Ortega-Escobar 2002), eye sensitivity (Yamashita & Nakamura 1999), and boldness (Jones et al. 2011).

Along with coordinating internal events (Green et al. 2008), it is widely believed that a selective advantage to having a circadian clock is that it allows organisms to anticipate, rather than simply respond to, regular changes in their environment associated with the 24 h solar day (for review see: Yerushalmi & Green 2009). Theory predicts that, the more closely the period of the circadian clock resonates with the 24 h solar day, the more effectively it can be used to regulate physiology and behavior to align with, or anticipate, daily environmental changes. Evidence consistent with this prediction is that dissonance between the period of the circadian clock and that of the solar day leads to reduced fitness in cyanobacteria (Woelfle et al. 2004) and mice (Spoelstra et al. 2016), as well as adverse physiological consequences in humans (Scheer et al. 2009). It was, therefore, surprising to discover that a trashline orbweaver *Cyclosa turbinata* (Walckenaer, 1841) (Araneidae) has a mean circadian free-running period (FRP) of 18.5 h (Moore et al. 2016), representing the shortest known naturally occurring circadian period in animals. This period length is similar to laboratory-generated mutants in *Drosophila* (Ralph & Menaker 1988) and hamsters (Konopka & Benzer 1971; Monecke et al. 2011). Another study of the orbweaver *Metazygia wittfeldae* (McCook, 1894) (Araneidae), found a mean FRP of 22.7 h, including two individuals with periods shorter than 20 h (Jones et al. 2018). There is evidence that having an internal clock period different from 24 h facilitates entrainment to the solar day (reviewed in Johnson et al. 2003, and discussed below). There is also natural variation in FRP among species, which can be amplified by environmental conditions (reviewed in Aschoff 1979, and discussed below). However, the vast majority of measured (non-mutant) FRPs are within 1 h of 24 h, suggesting that spiders may be quite unusual.

In addition to the variation in FRP among spider species, there is also an unusual amount of variation in FRP within species. In *C. turbinata,* the standard error of the mean (SEM) in FRP was 0.13 h, and the range among the studied individuals was 15.7 – 27.2 h (Moore et al. 2016). For *M. wittfeldae,* the standard error of the mean for FRP was 0.24 h, with endogenous period ranging from 18.75 to 23.8 h (Jones et al. 2018). Other spider species show similar or greater variation in FRP (Moore and Jones unpublished data) including *Parasteatoda tepidariorum* (C. L. Koch, 1841) females (Mah et al. 2018). It is a challenge to reconcile these high levels of variation in FRP with the general consensus that FRP should be under strong selection to resonate with the 24-h day (Yerushalmi & Green 2009). If FRP in spiders is a fixed inherited trait, then the observed variation would reflect among-individual genetic differences. Thus, such wide variation in FRP would suggest that it is *not* under strong selection to resonate with the solar day in spiders. A non-mutually exclusive alternative is that this variation may be the result of plasticity in the period of the clock, perhaps reflecting differences among individuals' age, sex, foraging success, or other life experiences (Aschoff 1979; Shemesh et al. 2007; Dubruille & Emery 2008). This plasticity hypothesis has added plausibility from the fact that most of the spider studies cited above used wild-collected individuals of unknown previous experience. Because of their accessibility, diversity of life history strategies, and distinct behaviors, spiders have great potential to contribute to our understanding of the mechanisms, ecology, and evolution of circadian rhythms. In fact, there is a recent push for such integration of physiology-focused chronobiology with ecology (Schwartz et al. 2017; Helm et al. 2017). Our broad objective with this study is to deepen our understanding of the nature of circadian rhythms and their variation within and among species of spiders, a group which could be a model system for integrating chronobiology and ecology. Specifically, we are beginning to address the question of how an individual's sex affects circadian rhythms in spiders. To our knowledge, all studies of FRP in spiders have been on females (Seyfarth 1980; Suter 1993; Ortega-Escobar 2002; Moore et al. 2016; Jones et al. 2018), including *P. tepidariorum* (Mah et al. 2018), or unidentified sex sub-adults (Soriano-Morales et al. 2013). In this study we present an analysis of diel and circadian patterns of locomotor activity in wild-collected male *P. tepidariorum*.

METHODS

Study species.—*Parasteatoda tepidariorum* (Theridiidae), the common house spider, occurs widely in North America, and is commonly found around human structures (Bradley 2013). Of particular importance to this study is that this species captures both crawling and flying prey at a relatively constant rate throughout the day and night (Riechert & Cady 1983). Both male and female juveniles disperse from their natal webs and begin foraging individually from the second instar to maturity (Valerio 1977). Whereas females remain in their own webs indefinitely, males abandon their webs upon maturing to search for mating opportunities, and are frequently found in the webs of penultimate instar females (Ewing 1918). Male spiders for this study were collected in June 2018 from the exteriors of multiple inhabited structures and outbuildings around Washington County, Tennessee, USA, and placed into the experiment within two days after collection. The spiders were all adults, though specific age, mating status, and exposure to artificial lights were unknown.

Locomotor activity.—To record locomotor activity, individuals ($n = 19$) were placed in 25 mm diameter X 100 mm length, clear glass tubes loosely capped to allow airflow, which were then inserted into a locomotor activity monitor (model LAM25, Trikinetics Inc.,Waltham, Massachusetts). During recording, the monitors were kept in a temperature-controlled environmental chamber (25 ± 0.5 °C) under a light-dark cycle containing 12 hours of light and 12 hours of dark (LD 12:12) for four days followed by eight days of constant darkness (DD). Lights-on occurred at 08:00 h and lights-off at 20:00 h. Light during photophase was provided by two horizontally mounted, 32 W fluorescent tubes and the illuminance was 1400-1600 lux at the level of the activity monitor. Open containers of water placed in the chambers maintained humidity during the experiment. We recorded activity bouts (measured by crossings of infrared beams bisecting the long axis of the tubes) in one-minute bins. The results were analyzed using Clocklab Analysis 6 Software (Actimetrics, Wilmette, IL, U.S.A.), testing for significant periodicity under DD conditions (i.e., free-running periods, FRPs) using Lomb-Scargle periodogram analyses, the method of choice to analyze records with frequent or large gaps (Van Dongen et al. 1999). To be conservative in our analyses, we chose the most stringent P value of 0.001 in determining significant periodicities.

RESULTS

Entrainment profiles.—All of the 19 individuals we tested showed locomotor activity during the LD 12:12 cycle, but one individual died shortly into the DD phase and was omitted from the analyses. The mean phase angle of entrainment (the timing of a biological event relative to an external time cue) for the onset of activity in LD 12:12 was 0.55 ± 0.52 h (mean ± SD here and throughout results section) after lights off. We observed no differences in the entrainment profiles among the four days of LD12:12, so an overall average profile is presented (Fig. 1). A clear pattern in the entrainment profile is a ramping up of activity within the first 30–40 minutes immediately following lights-off. In most individuals, there was a large bout of locomotor activity in early scotophase, followed by irregularly scattered, diminishing activity through the rest of scotophase (observed as a steady decline in the averaged entrainment profile). There also was typically a small burst of activity immediately after lights on. Because these events occur in LD but do not persist in DD, they are often interpreted as an exogenous effect (Englemann & Mack 1978). We quantified the diurnality/nocturnality of activity in LD 12:12 using a modification of the 'diurnality index' (Hoogenboom et al. 1984) finding that activity was almost exclusively nocturnal with a mean DiNoc ratio ($\text{Activity}_{\text{light}} - \text{Activity}_{\text{dark}} / \text{Activity}_{\text{total}}$; Suter & Benson 2014) of -0.88 ± 0.12.

Circadian free-runs.—Under constant darkness, all but one of the 19 individuals showed free-running locomotor activity with Lomb-Scargle significance at $P < 0.001$. The FRPs were generally short, with a mean of 21.18 ± 1.5 h (Fig. 2). The distribution of FRPs best fit a 2-parameter exponential distribution (Anderson-Darling test statistic = 0.32, $P > 0.25$), with most falling between 19.5 and 21.5 h, but with three closer to 24 h (Fig. 3). Also, in four of the individuals it appeared that the FRP was lengthening over the course of DD, appearing as curvature in the actogram (an example is seen in Fig. 2b). Along with the primary FRP peak on the periodograms, all showed significant secondary peaks (Figs. 2 a–d). The only obvious generalization in these secondary peaks was that there tended to be one at close to half the period of the FRP.

Of particular note in the actograms was that the onset of activity in DD usually did not extrapolate back to the last onset of LD, but rather to mid-to-late photophase (indicated by open arrows, Figs. 2 a–c). In fact, the mean onset of activity on the 1st day of DD was 13.73 ± 3.4 h, an average of 6.82 ± 3.3 h earlier than individuals' phase angle of entrainment under LD 12:12. This was generally consistent among individuals with one exception for which the initial onset of activity under DD was actually later than that observed under LD (Fig. 2d). To confirm that the phase advancement going into the first day of DD was more than just the effect of a short FRP, we compared the phase advancement between the last day of LD and the first day of DD (mean 7.46 ± 2.22 h) with the phase advancement between day 1 and day 2 of DD (mean 5.24 ± 1.83 h). The two-phase advancements were

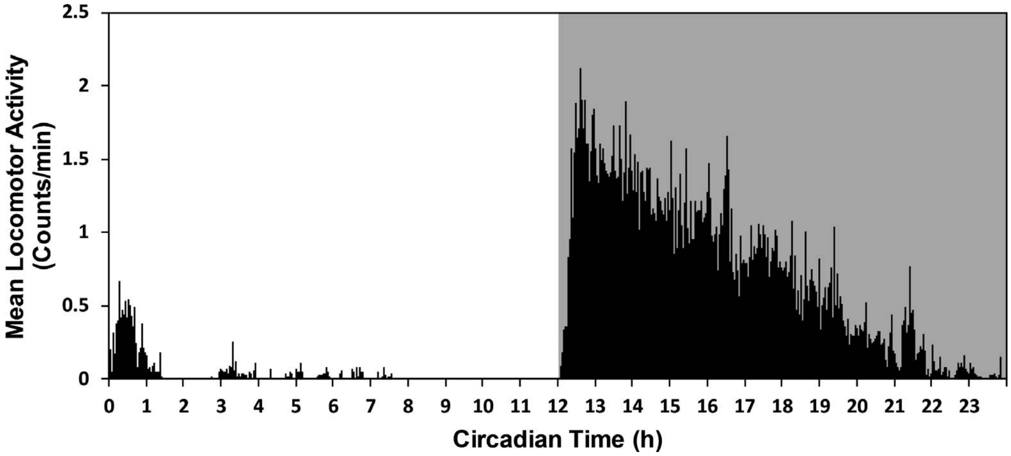

Figure 1.—Pattern of entrainment to LD 12:12 h cycle. Shown are mean activity levels, in 1-min bins, for five consecutive days of exposure to the light-dark cycle. Shading indicates scotophase.

statistically different (paired T-test: t = 2.93, df = 13, P = 0.012), with the phase advancement in the transition from LD to DD being about 2.2 h greater than the following day.

DISCUSSION

Our results here corroborate recent studies demonstrating that spiders, from a chronobiological perspective, are consistently inconsistent with well-studied model species, validating the recent call to phylogenetically diversify the discipline (Schwartz et al. 2017; Helm et al. 2017). In the case of male *P. tepidariorum*, both the mean and variation in FRP are noteworthy. The mean FRP of 21.18 h, though not as short as that in *C. turbinata* (Moore et al. 2016), is nearly three hours shorter than the solar day. Therefore, these animals must phase-delay about three hours per day simply to remain entrained. Theory (reviewed by Johnson et al. 2003) suggests that having a circadian period different from 24 h actually benefits entrainment to the solar day in terms of maintaining a stable phase angle of entrainment (Pittendrigh & Daan 1976) and in adapting to changes in day length (Daan & Aschoff 1975). Models and evidence, however, also suggest that entrainment becomes increasingly difficult or impossible if the difference between the circadian clock and external light cycle is greater than one hour (Johnson et al. 2003). Moreover, evidence broadly suggests that circadian dissonance greater than an hour has myriad adverse physiological consequences (reviewed in Evans & Davidson 2013). In spite of this, we now know of three spider species, representing two families, that are over an hour out of resonance with the solar day: *P. tepidariorum* (this study), *C. turbinata* (Moore et al. 2016), and *M. wittfeldae* (Jones et al. 2018). This strongly suggests that spiders are 'released' from such adverse physiological consequences, relaxing the selection to resonate with the solar day. As yet, we have no insight into the mechanism of this release, nor do we have evidence to distinguish if the release is allowing FRP to selectively adapt or merely drift. However, the consistently wide variation in within-species FRP observed here (SEM: ± 0.38 h) and elsewhere (*C. turbinata* SEM: ± 0.13 h, Moore et al. 2016; *M. wittfeldae* SEM: ± 0.24 h, Jones et al. 2018; *Lycosa tarantula* Linnaeus, 1758 SEM: ± 0.59 h, Ortega-Escobar 2002; *Cupiennius salei* (Keyserling, 1877)(Trechaleidae) SEM ± 0.31 h, Seyfarth 1980) would seem inconsistent with the idea that species are under strong purifying selection for a particular FRP. For reference, typical within-species standard error of the mean in FRP is much less, at about ± 0.03 h in humans (Czeisler et al. 1999), ± 0.07 h in golden hamsters, (Ralph & Menaker 1988), and only ± ~ 0.02 h in *D. melanogaster* (Seggio 2013). We should note that environmental conditions, particularly those involving light, can have lasting effects on an individual's FRP (Aschoff 1979). The animals in the above studies used lab-reared animals, whereas animals in this study were collected haphazardly from around buildings. Thus, differential exposure to artificial light may have influenced the variation among individuals. Further study of this possibility is certainly warranted.

The presence of significant periodicities at approximately half the length of the FRP observed under DD in this study is similar to the phenomenon observed in *M. wittfeldae*, but in *M. wittfeldae* the peaks were more precisely half the FRP (Jones et al. 2018). While not well understood, other studies have attributed activity bouts at a phase angle juxtaposed by 180° to the main activity bout, but free-running with the same period, to separate outputs of one circadian oscillator (Wiedenmann 1980). In *M. wittfeldae*, these peaks were quite pronounced in the periodograms and corresponded to a distinct bout of activity in the actograms (Jones et al. 2018), whereas in male *P. tepidariorum*, it is not clear what component of the actogram corresponds to the second periodogram peaks.

An aspect of the FRPs that has not been reported in other spider species, including *C. turbinata* with its unusually short FRP (Moore et al. 2016), is that the onset of activity in DD does not extrapolate back to the last onset in LD, but rather about seven hours earlier to mid-photophase. This would suggest that the circadian phase angle of entrainment for onset of activity in LD is actually much earlier than the observed phase angle, but that locomotor activity governed by the circadian clock is masked in the presence of light (for review of masking see: Rietveld et al. 2011) but revealed during constant dark conditions. An alternative explanation for the misalignment would be that the lack of light in DD on exclusively the

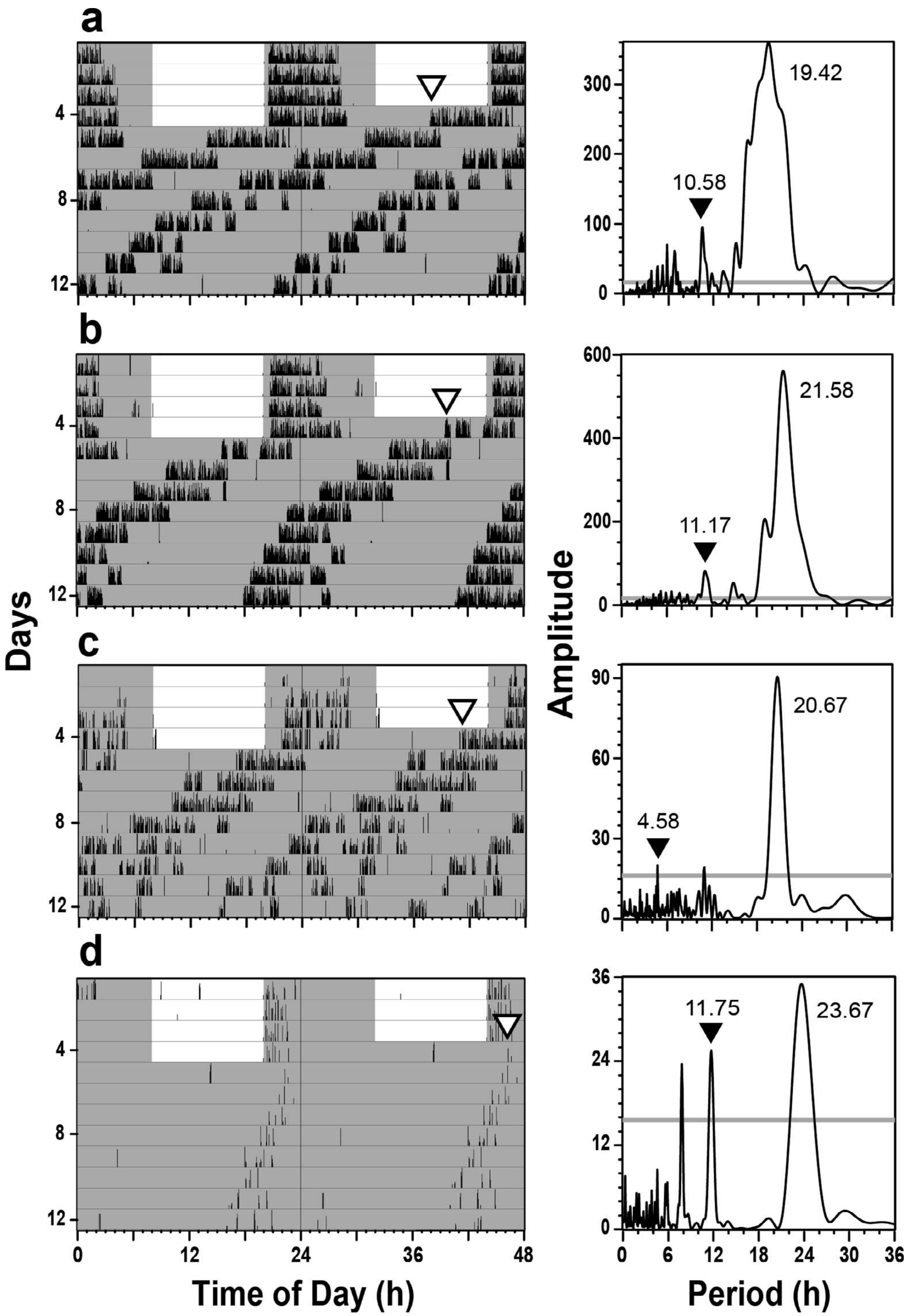

Figure 2.—Double-plotted actograms, each with its accompanying periodogram, of four individuals (a–d) illustrating entrainment and free-running locomotor activity in four different males of *P. tepidariorum*. Actograms (left panels) depict the timing of activity for four consecutive days under a LD 12:12 h cycle, followed by constant dark (DD) conditions thereafter. Dark periods are represented by gray background. Lomb-Scargle periodograms (right panels) indicate significant periodicities ($P < 0.001$) of locomotor activity under DD. Filled triangles on the periodograms indicate the strongest secondary peak in the data, while open triangles in the actograms indicate the point to which the free-running activity extrapolates back to on the last day of LD.

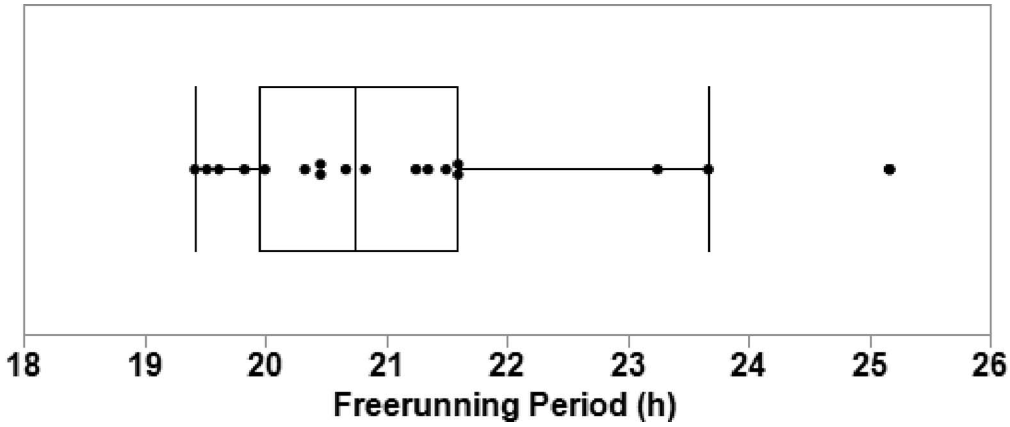

Figure 3.—Frequency distribution of circadian free-running periods of locomotor activity under DD conditions for *P. tepidariorum* males. Individual FRPs are indicated as open circles, the mean FRP is indicated by filled triangle, and the dashed line indicates the 'expected' FRP of 24 h.

first subjective day causes a one-time circadian phase advance of nearly seven hours. The latter hypothesis seems less likely, but further experiments are needed to confirm masking of locomotor activity by light.

In chronobiological comparisons between females and males in insects, differences are often observed in entrainment profiles (Loher 1979; Rivas et al. 2008; Bertossa et al. 2013; Prohaska et al. 2018). Two studies of dipteran insects found that, along with differences in entrainment profiles, males had significantly shorter FRPs (Helfrich-Förster 2000; Rund et al. 2012). Mah et al. (2018) provide a description of diel and circadian locomotor activity in female *P. tepidariorum*, finding very similar patterns to what we observed here in males. As shown in the present study with males, there is a sharp increase in activity immediately after lights-off followed by a steady decline through the night. The only consistent difference was that females showed more daytime activity, which is reflected in the average DiNoc ratios (mean ± SD: females: -0.71 ± 0.05; males: -0.88 ± 0.12). The fact that males are more strictly nocturnal might be advantageous because at maturity they leave the relative safety of their webs in search of females (Ewing 1918). The FRPs are statistically indistinguishable between females and males (Mean ± SD, females: 21.7 ± 1.12 h; males 21.18 ± 1.5 h; $T = 1.11$, $df = 32$, $P = 0.14$), and both show the phenomenon of the onset of activity in DD not extrapolating back to the last onset in LD. Notably, there is high within-species variation in FRP for both males and females. While far from distinguishing whether this variation reflects phenotypic plasticity or underlying genetic differences, the latter might seem more supported given that, in all likelihood, males and females experience different ecological conditions. We are currently exploring the contribution of genetics and phenotypic plasticity in lab-reared clutches of *P. tepidariorum*.

Returning to the misalignment of activity onset in DD with LD: if *P. tepidariorum* phase angle of entrainment for locomotor activity onset is in the mid to late photophase but hidden by masking, why might this be so? We speculate that this phenomenon might cause individuals to be 'neurologically primed' in late photophase, allowing them to capture prey opportunistically should environmental conditions permit. There is circumstantial evidence to support this. It is widely believed that predation risk for web-building spiders is generally higher during the day because of the presence of visual predators such as birds and wasps (Foelix 2011). Furthermore, day-hunting mud-dauber wasps are a particular threat to *P. tepidariorum* (Muma & Jeffers 1945). Also, spiders are precariously balanced as both predators and prey, and their own foraging behavior can increase their predation risk (Riechert & Hedrick 1993; Pruitt et al. 2008; Watts et al. 2014). Taken together, this would suggest that masking locomotor activity in the light would reduce predation risk (assuming a correlation of locomotor activity and likelihood of attacking prey as seen in another spider; Watts et al. 2014). Though not tested in *P. tepidariorum* habitat specifically, two studies found that spider prey abundance is highest in the late afternoon to early evening, and lowest at dawn (Watts et al. 2014, 2017). This would suggest that the opportunity cost of not foraging in the day is greater in the late afternoon than early morning. Finally, because *P. tepidariorum* tend to build webs under rock outcrops (Riechert & Cady 1983), individuals likely experience higher variation in the onset of local darkness relative to more exposed species (due to depth and angle of rock cover, cloudiness, etc). This would suggest a particular advantage of being able to respond to late-day opportunities when it is dark enough in *P. tepidariorum* (and other secluded species) compared to exposed species. Admittedly, all this is speculation, but it does demonstrate how spiders may be an ideal system for future studies integrating classic chronobiology and ecology (Schwartz et al. 2017; Helm et al. 2017).

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LITERATURE CITED

Aschoff, J. 1979. Circadian rhythms: influences of internal and external factors on the period measured in constant conditions. Ethology 49:225–249.

Bertossa, R.C., J. van Dijk, W. Diao, D. Saunders, L.W. Beukeboom

& D.G.M. Beersma. 2013. Circadian rhythms differ between sexes and closely related species of *Nasonia* wasps. PLoS ONE 8(3): e60167.

Bradley, R.A. 2013. Common Spiders of North America. University of California Press, Los Angeles.

Czeisler, C.A., J.F. Duffy, T.L. Shanahan, E.N. Brown, J.F. Mitchell, D.W. Rimmer et al. 1999. Stability, precision, and near-24-hour period of the human circadian pacemaker. Science 284:2177–2188.

Daan, S. & J. Aschoff. 1975. Circadian rhythms of locomotor activity in captive birds and mammals: heir variations with season and latitude. Oecologia 18:269–290.

Dubruille, R. & P. Emery. 2008. A plastic clock: how circadian rhythms respond to environmental cues in *Drosophila*. Molecular Neurobiology 38:129–145.

Engelmann, W. & J. Mack. 1978 Different oscillators control the circadian rhythm of eclosion and activity in *Drosophila*. Journal Comparative Physiology 127:229–237.

Evans, J.A. & A.J. Davidson. 2013. Chapter ten: health consequences of circadian disruption in humans and animal models. Progress in Molecular Biology and Translational Science 119:283–323.

Ewing, H.E. 1918. The life and behavior of the house spider. Proceedings of the Iowa Academy of Science 25:177–204.

Foelix R.F. 2011. Biology of Spiders, 3rd Edition. Oxford University Press. New York.

Green, C.B., J.S. Takahashi & J. Bass. 2008. The meter of metabolism. Cell 134:728–742.

Helfrich-Förster, C. 2000. Differential control of morning and evening components in the activity rhythm of *Drosophila melanogaster*—Sex-specific differences suggest a different quality of activity. Journal of Biological Rhythms 15:135–154.

Helm, B., M.E. Visser, W. Schwartz, N. Kronfeld-Schor, M. Gerkema, T. Piersma et al. 2017. Two sides of a coin: ecological and chronobiological perspectives of timing in the wild. Philosophical Transactions of the Royal Society B 372: 20160246

Hoogenboom, I., S. Daan, J.H. Dallinga & M. Schoenmakers. 1984. Seasonal change in the daily timing of behaviour of the common vole, *Microtus arvalis*. Oecologia 61:18–31.

Johnson, C.H. & T. Kondo. 2001. Circadian rhythms in unicellular organisms. Pp. 61–78. *In* Circadian Clocks, Volume 12. (J.S. Takahashi, F.W. Turek & R.Y. Moore, eds.). Kluwer Academic/ Plenum Publishers, New York.

Johnson, C.H., J.A. Elliot & R. Foster. 2003. Entrainment of circadian programs. Chronobiology International 5:741–774.

Jones, T.C., T.S. Akoury, C.K. Hauser & D. Moore. 2011. Evidence of circadian rhythm in antipredator behaviour in the orb-weaving spider *Larinioides cornutus*. Animal Behaviour 82:549–555.

Jones T.C., R.J. Wilson & D. Moore. 2018. Circadian rhythms of locomotor activity in *Metazygia wittfeldae* (Araneae: Araneidae). Journal of Arachnology 46:26–30.

Konopka, R.J. & S. Benzer. 1971. Clock mutants of *Drosophila melanogaster*. Proceedings of the National Academy of Sciences U.S.A. 68:2112–2116.

Loher, W. 1979. Circadian rhythmicity of locomotor behavior and oviposition in female *Teleogryllus commodus*. Behavioral Ecology and Sociobiology 5:253–262.

Mah A., N. Ayoub, D. Moore, N. Toporikova & T.C. Jones. 2018. Computational models of spider activity patterns integrated with behavioral experiments hypothesize adaptive benefits of the circadian clock. Thesis, online at: https://repository.wlu.edu/bitstream/handle/11021/34112/RG38_Mah_NEUR_2018.pdf?sequence=1&isAllowed=y

Monecke, S., J.M. Brewer, S. Krug & E.L. Bittman. 2011. Duper: A mutation that shortens hamster circadian period. Journal of Biological Rhythms 26:283–292.

Moore, D., J.C. Watts, A. Herrig & T.C. Jones. 2016. Exceptionally short-period circadian clock in *Cyclosa turbinata*: regulation of locomotor and web-building behavior in an orb-weaving spider. Journal of Arachnology 44:388–396.

Muma, M.H. & W.F. Jeffers. 1945. Studies of the prey of several mud-dauber wasps. Annals of the Ecological Society of America 38:245–255.

Ortega-Escobar, J. 2002. Circadian rhythms of locomotor activity in *Lycosa tarantula* (Araneae, Lycosidae) and the pathways of ocular entrainment. Biological Rhythms Research 33:561–576.

Panda, S., J.B. Hogenesch & S.A. Kay. 2002. Circadian rhythms from flies to human. Nature 417:329–335.

Pittendrigh, C.S. 1960. Circadian rhythms and the circadian organization of living systems. Pp. 159–184. *In* Cold Spring Harbor Symposia on Quantitative Biology: Biological Clocks. Long Island Biological Association, New York.

Pittendrigh, C.S. & S. Daan. 1976. A functional analysis of circadian pacemakers in small rodents. IV. Entrainment: pacemaker as a clock. Journal of Comparative Physiology 106:291–331.

Prohaska, F., K.H. Joplin & D. Moore. 2018. Effects of gender, age, and nutrition on circadian locomotor activity rhythms in the flesh fly *Sarcophaga crassipalpis*. Journal of Insect Physiology 107:265–275.

Pruitt, J.N., S.E. Riechert & T.C. Jones. 2008. Behavioural syndromes and their fitness consequences in a socially polymorphic spider, *Anelosimus studiosus*. Behavioral Ecology 76:871–879.

Ralph, M.R. & M. Menaker. 1988. A mutation of the circadian system in golden hamsters. Science 241:1225–1227.

Riechert, S.E. & A.B. Cady. 1983. Patterns of resource use and tests for competitive release in a spider community. Ecology 64:899–913.

Riechert, S.E. & A.V. Hedrick. 1993. A test for correlations among fitness-linked behavioural traits in the spider *Agelenopsis aperta* (Araneae: Agelenidae). Animal Behaviour 46:669–675.

Rietveld, W.J., D.S. Minors & J.M. Waterhouse. 2011. Circadian rhythms and masking: an overview. Chronobiology International 10:306–312.

Rivas, G.B.S., N.A. Souza & A.A. Peixoto. 2008. Analysis of the activity patterns of two sympatric sandfly siblings of the *Lutzomyia longipalpis* species complex from Brazil. Medical and Veterinary Entomology 22:288–290.

Rund, S.S.C., S.J. Lee, B.R. Bush & G.E. Duffield. 2012. Strain- and sex-specific differences in daily flight activity and the circadian clock of *Anopheles gambiae* mosquitoes. Journal of Insect Physiology 58:1609–1619.

Scheer, F.A.J.L., M.F. Hilton, C.S. Mantzoros & S.A. Shea. 2009. Adverse metabolic and cardiovascular consequences of circadian misalignment. Proceedings of the National Academy of Sciences 106:4453–4458.

Schwartz, W.J., B. Helm & M.P. Gerkema. 2017. Wild clocks: preface and glossary. Philosophical Transactions of the Royal Society B 372: 20170211.

Seggio, J.A. 2013. The free-running period of *Drosophila melanogaster* is not affected by the length of the tube in a *Drosophila* Activity Monitor (DAM). Drosophila Information Service 96:197–199.

Seyfarth, E.A. 1980. Daily patterns of locomotor activity in a wandering spider. Physiological Entomology 5:199–206.

Sheer F.A.J.L., M.F. Hilton, C.S. Mantzoros & S.A. Shea. 2009. Adverse metabolic and cardiovascular consequences of circadian misalignment. Proceedings of the National Academy of Sciences 106:4453–4458.

Shemesh, Y., M. Cohen & G. Bloch. 2007. Natural plasticity in circadian rhythms is mediated by reorganization in the molecular clockwork in honeybees. FASEB Journal 21:2304–2311.

Soriano-Morales, S., O. Caballero-Hernández, M. Dávila-Montes, J.B. Morales-Malacara & M. Miranda-Anaya. 2013. Circadian locomotor activity and entrainment by light cycles in cave spiders

(Dipluridae and Ctenidae) at the cave Los Riscos, Qro. México. Biological Rhythm Research 44:949–955.

Spoelstra, K., M. Wikelski, S. Daan, A.S.I. Loudon & M. Hau. 2016. Natural selection against a circadian clock gene mutation in mice. Proceedings of the National Academy of Sciences, USA 113:686–691.

Suter, R.B. 1993. Circadian rhythmicity and other patterns of spontaneous motor activity in *Frontinella pyramitela* (Linyphiidae) and *Argyrodes trigonum* (Theridiidae). Journal of Arachnology 21:6–22.

Suter, R.B. & K. Benson. 2014. Nocturnal, diurnal, crepuscular: activity assessments of Pisauridae and Lycosidae. Journal of Arachnology 42:178–191.

Valerio, C.E. 1977. Population structure in the spider *Achaearanea tepidariorum* (Araneae, Theridiidae). Journal of Arachnology 3:185–190.

Van Dongen, H.P.A., E. Olofsen, J.H. Van Hartevelt & E.W. Kruyt. 1999. A procedure for multiple period searching in unequally spaced time-series with the Lomb-Scargle method. Biological Rhythm Research 30:149–177.

Watts J.C., A. Herrig, W.D. Allen & T.C. Jones. 2014. Diel patterns of foraging aggression and antipredator behaviour in the trashline orb-weaving spider, *Cyclosa turbinata*. Animal Behaviour 94:79–86.

Watts J.C., M. Miller, A. Herrig, T.C. Jones & B. Tenhumberg. 2017. Temporal variation in predation risk may explain daily rhythms of foraging behavior in an orb-weaving spider. American Naturalist 191:74–87.

Wiedenmann, G. 1980. Two peaks in the activity rhythm of cockroaches controlled by one circadian pacemaker. Journal of Comparative Physiology 137:249–254.

Woelfle, M.A., Y. Ouyang, K. Phanvijhitsiri & C.H. Johnson. 2004. The adaptive value of circadian clocks: an experimental assessment in cyanobacteria. Current Biology 14:1481–1486.

Yamashita, S. & T. Nakamura. 1999. Circadian oscillation of sensitivity of spider eyes: diurnal and nocturnal spiders. Journal of Experimental Biology 202:2539–2542.

Yerushalmi S. & R.M. Green. 2009. Evidence for adaptive significance of circadian rhythms. Ecology Letters 12:970–981.

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