

## Article

# Daily activity rhythms, chronotypes, and risk-taking behavior in the signal crayfish

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## Abstract

Consistent inter-individual differences in daily activity rhythms (i.e., chronotypes) can have ecological consequences in determining access to food resources and avoidance of predators. The most common measure to characterize chronotypes in animals as well as humans is the onset of activity (i.e., early or late chronotypes). However, daily activity rhythms may also differ in the relative amount of activity displayed at particular time periods. Moreover, chronotypes may also be linked to other consistent inter-individual differences in behavior (i.e., personality), such as the propensity to take risks. Here, we used the signal crayfish *Pacifastacus leniusculus* to test the presence of chronotypes and risk-taking personality traits and a potential behavioral syndrome between these traits. We first exposed crayfish to 5 days of light–darkness to measure daily activity rhythms and then we applied a visual predator-simulating stimulus in 2 different contexts (neutral and food). Our results showed consistent (i.e., across 5 days) inter-individual differences in the relative nocturnal activity displayed in the early and middle, but not in the late part of darkness hours. Moreover, while crayfish displayed inter-individual differences in risk-taking behavior, these were not found to be consistent across 2 contexts. Therefore, we were not able to formally test a behavioral syndrome between these 2 traits. In conclusion, our study provides the first evidence of chronotypes in the relative amount of activity displayed at particular time periods. This could be a valuable information for applied ecological aspects related to the signal crayfish, which is a major invasive species of freshwater ecosystems.

**Key words:** behavioral syndrome, biological rhythms, boldness, feeding, locomotor activity

The behavior and physiology of almost all animal taxa is characterized by daily activity rhythms (Dunlap et al. 2004). Daily activity rhythms are usually synchronized to the day–night cycle, but other environmental or social factors can also play a role (Castillo-Ruiz et al. 2012; Hut et al. 2012). Daily behavioral rhythms provide ecological advantages with respect to exploitation of food resources and avoidance of predation (Kronfeld-Schor and Dayan 2003; Kronfeld-Schor et al. 2013; Helm et al. 2017). In particular, consistent inter-individual differences in daily activity rhythms—referred to

as chronotypes (Helm et al. 2017)—can have fitness consequences. For example, in the desert golden spiny mouse, *Acomys russatus*, individuals that arrive earlier to a foraging patch gain more food than individuals that arrive later (Levy et al. 2012), while Razorfish (*Xyrichtys novacula*) with early-active chronotypes could experience higher mortality from fisheries than late chronotypes (Martorell-Barceló et al. 2018). Daily activity rhythms may not only differ in the onset of activity (i.e., early active or late active), but also in the relative amount of activity displayed at particular time periods,

which can subsequently be linked to energy budget (Kronfeld-Schor and Dayan 2003; Helm et al. 2017). Therefore, consistent inter-individual differences in the relative amount of activity displayed at particular time periods could be a fitness-related trait (Kronfeld-Schor and Dayan 2003; Helm et al. 2017).

The degree of risk-taking behaviors (often referred to as “boldness”) such as locomotor activity in the presence of a predator or spatial distance to a predator, affects survival of an individual but may have consequences for access to food resources (Smith and Blumstein 2008; but see Moiron et al. 2020). According to the pace-of-life syndrome hypothesis (Réale et al. 2010), life-history, behavioral, and physiological traits are correlated along a fast-to-slow continuum. Fast life-history traits are expected to co-vary and co-evolve with an increase of risk-taking behavior and high metabolic rate. The reason is the acquisition of resources that needed to maintain a fast life history (i.e., rapid juvenile growth or high reproductive investment) favors risky behaviors such as feeding in the presence of predators. However, risk-taking behavior can also be evolutionarily linked to daily activity rhythms as demonstrated by recent studies. Indeed, Tudorache et al. (2018) showed that Zebrafish, *Danio rerio*, individuals displaying higher locomotion early in the activity period were also more risk-taking than individuals without clear peaks of early activity. Similarly, Sbragaglia et al. (2021) showed that Zebrafish displaying higher locomotion early in the activity period were also displaying more risk-taking behavior in an evolutionary context imposed by experimental size-selective harvesting. Therefore, it is plausible that a behavioral syndrome between risk-taking behavior and daily activity rhythms has an adaptive value.

We use the signal crayfish, *Pacifastacus leniusculus*, to test a relationship between daily activity rhythms and risk-taking behavior. Previous studies on crustaceans have shown repeatability in boldness as well as behavioral syndromes with other traits such as aggression (Briffa et al. 2008; Mowles et al. 2012). The noble crayfish, *Astacus astacus*, show repeatability in emergence across time and context (presence or absence of predator cues; Vainikka et al. 2011). Signal crayfish display correlation between boldness, aggression, and activity but the repeatability of these behaviors was not tested (Pintor et al. 2009). Moreover, previous studies showed that the signal crayfish is a nocturnal species (Nyström 2005; Edmonds et al. 2011; Thomas et al. 2016), but a clear daily activity pattern was never recorded for this major invasive species. Nocturnal activity of crayfish has been suggested as an adaptation to reduce susceptibility to diurnal predators (Holdich 2002), therefore it may be linked to risk-taking behavior.

Here, we test the hypothesis that daily locomotor activity and risk-taking behavior form a behavioral syndrome in the signal crayfish. We tackle the following research questions:

- i. do crayfish show consistent inter-individual differences in their relative nocturnal activity (early, middle, and late) across days, as indicative of chronotypes?
- ii. do crayfish show consistent inter-individual differences in risk-taking behavior across contexts (neutral and food), as indicative of personality traits? and
- iii. do crayfish show a link between mean relative daily nocturnal activity and risk-taking behavior, as indicative of a behavioral syndrome between these traits?

## Materials and Methods

### Sampling and acclimation

Crayfish were sourced from a commercial supplier using a farmed population (Flowers Farm Lakes, Dorchester, UK). All crayfish

arrived in the same shipment 2 weeks prior to start of the experimental trials. They were acclimated in communal tanks (60 × 45 × 25 cm) in sex-segregated groups of 10–15 individuals. The communal tanks included broken elements of clay flower pots large enough to provide shelter for crayfish. Crayfish were fed twice a week an alternating diet of cooked prawn and peas. Feeding was targeted toward individuals (3 peas or 1 prawn) and sometimes resulted in conspecific fights. Aggressive interaction settled soon after all animals had received food, but nocturnal competition over shelters may have resulted in unknown fight histories of individuals (Fero et al. 2007). The light–darkness cycle was 12–12 h and water temperature was kept at  $15 \pm 1^\circ\text{C}$ .

We used a total of 24 male crayfish (cephalothorax length:  $36.0 \pm 2.7$  mm). We conducted a total of three experimental trials from May to July. We ran each trial with 8 randomly selected crayfish in an experimental room where light (light–darkness cycle of 12–12 h) and water temperature ( $15 \pm 1^\circ\text{C}$ ) were controlled. Light was provided by fluorescent lamps at an intensity of 0.25 Klux. During the darkness period dim red light (0.01 Klux) was provided by monochromatic light-emitting diodes to allow video recording.

The experimental setup consisted of 8 separated glass aquaria (45 cm × 30 cm × 25 cm). We equipped each tank with a recirculation pump (provided with a filter). We glued white opaque cloth to the bottom of the tank to create a suitable surface for the crayfish to walk on and the appropriate background to apply video imaging analysis (see below). We placed a shelter (12 cm long) in each tank, cutting transversally a polyvinyl chloride pipe of a diameter of 8 cm. We covered the sides of each tank with white fabric to eliminate any visual disturbance from neighboring tanks and we placed the tanks behind light proof curtains to eliminate any kind of light contamination.

### Experimental design

We used an experimental assay divided in two phases. In the first phase, we left the crayfish undisturbed for five days recording a time-lapse video (10 s frequency) that we used to track daily locomotor activity rhythms (see below). During this phase, crayfish had always food available (prawns or peas as used in the acclimation period) that was replaced every day. In the second phase, we exposed crayfish to a scare stimulus in two different contexts (neutral and food contexts) to evaluate their level of risk-taking behavior. One of the most important antipredatory behaviors of crayfish includes reduced movement (Stein and Magnuson 1976). Therefore, we considered the amount of locomotor activity performed 30 min after the scare stimulus as a measure of risk-taking behavior (i.e., the less the locomotor activity the less risk-taking behavior). We applied the scare stimulus 2 days apart within 6 days after the locomotor activity recording (first stage of the experiment) moving a plastic black board (15 cm × 15 cm) forward and back 2 times over the surface (see [Supplementary Video S1](#)). During the neutral context nothing was changed in the experimental tank (food was not available), while during the food context a small piece of prawn was added after light-off (in this second phase of the experiment, crayfish were only fed for measuring risk-taking behavior in this context). The scare stimulus was applied within 2 h after light-on only when the crayfish was moving or approaching the food (displacement for more than 1 body length), otherwise the stimulus was not applied and the behavior scored with a 0 because our interpretation is that remaining in the shelter implies a low level of risk-taking behavior (Jurcak et al. 2016).

## Behavioral tracking

We quantified locomotor activity using automated video image analysis. Four universal serial usb webcams (1 webcam for 2 aquaria) were placed behind light-proof curtains on the top of the aquaria. We managed frame acquisition using open source camera security software (ispy: <http://www.ispyconnect.com>). The automated video image analysis was performed in Matlab 7.1 (The MathWorks, Natick, MA, USA), adapting a compilation of a script with the image processing toolbox previously used in several studies with decapods crustaceans (Sbragaglia et al. 2013, 2015a, 2015b). Briefly, the behavioral tracking was performed by means of algebraic subtraction of each image with a starting background image. Threshold filtering has been applied for removing noise and errors during tracking. The centroid coordinates of the tracked pixel matrix were used as position of the crayfish. When the crayfish was not recorded (mainly when it was inside the shelter), the last tracked position was kept until a new detection. The final output was a time series of the movement of each crayfish (cm) at a frequency of 10 s.

## Data treatment and statistical analysis

The 5-day time series of locomotor activity were analyzed using a Chi-square periodogram (Sokolove and Bushell 1978) to scan for periodicity and the percentage of variance (%V) was reported as a measure of rhythms' robustness (Refinetti 2006). Crayfish were considered rhythmic only in the case in which periodicity was statistically significant ( $P < 0.05$ ), otherwise crayfish were defined arrhythmic. Percentage of locomotor activity during darkness and mean overall activity across 5 days (i.e., the midline estimating statistic of rhythm that is represented as a horizontal threshold in waveform plots) were used to further characterize daily locomotor activity rhythms (Refinetti 2006). We also calculated the mean nocturnal activity (mean level of locomotor activity during darkness across 5 days). Relative nocturnal activity (early, middle, and late) was calculated for each of the 5 days as the proportion between locomotor activity in the 4-h periods (early: from light-off to 4 h after light-off; middle: from 4 h after light-off to 4 h before light-on; late: from 4 h before light-on to light-on) and the total activity performed during that day (i.e., in 24 h; Figure 1).

Repeatability was calculated across time (5 days) for daily relative nocturnal activity (early, middle, late; research question I) and across contexts (neutral and food; research question II) for risk-taking behavior (Nakagawa and Schielzeth 2010). The values of relative nocturnal activity (early, middle, and late) were transformed by finding the exponent ( $\lambda$ ), which made the values of the response variable as normally distributed as possible with a power transformation and a Gaussian distribution of error has been used to calculate repeatability. The values of risk-taking behavior were not transformed and a Poisson distribution was used to calculate repeatability for risk-taking behavior across context (neutral and food). Model fitting was examined by checking normality of residuals and plotting theoretical quantiles versus standardized residuals. The correlation between mean relative nocturnal activity and risk-taking behavior (research question III) was calculated with the non-parametric Kendall's coefficient of concordance followed by the estimation of Vargha and Delaney's A effect sizes (Vargha and Delaney 2000). Furthermore, we also calculated the correlation between mean relative nocturnal activity (early, middle, and late) and the midline estimating statistic of rhythm in order to assess whether the chronotypes were somehow related to mean levels of activity of individuals.

Analysis was performed using the software Eltemps ([www.eltemps.com](http://www.eltemps.com)) and R 3.3.1 (<https://www.R-project.org/>) with the following additional packages: "rcompanion" (<https://CRAN.R-project.org/package=rcompanion>), "rptR" (Stoffel and others 2017). In all cases, we used a 95% confidence interval.

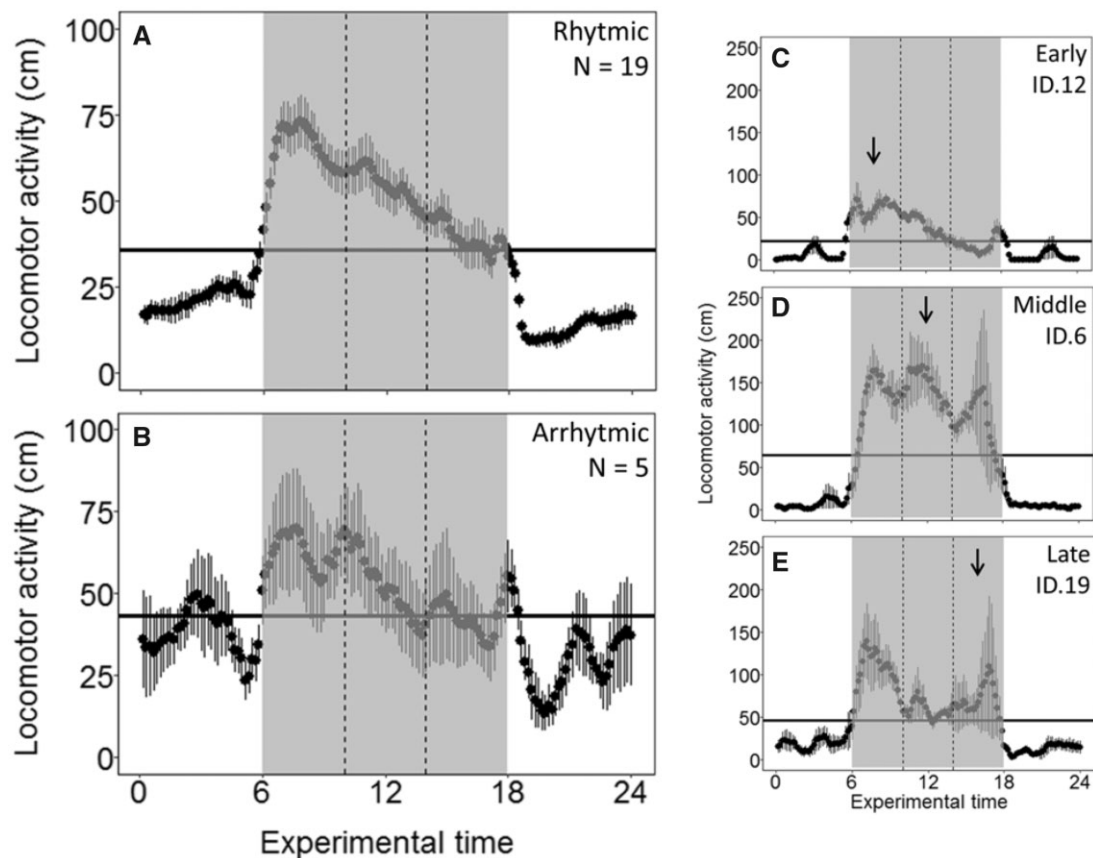
## Results

Time series analysis of the first stage of the experiment indicated robust daily locomotor activity rhythms (mean robustness  $\pm$  standard deviation =  $46 \pm 12\%$ ;  $n = 19$ ; 5 crayfish did not show significant periodicity in their activity rhythms) with more activity during darkness ( $72\% \pm 12\%$ ;  $n = 24$ ; Figure 1). Relative early and middle nocturnal activity were repeatable across days (early:  $R = 0.23$  [0.05–0.41];  $P < 0.01$ ; middle:  $R = 0.22$  [0.02–0.40];  $P < 0.01$ ;  $n = 24$ ; Figure 2A–B). In contrast, relative late nocturnal activity was not repeatable across days ( $R = 0.04$  [0–0.19];  $P = 0.324$ ;  $n = 24$ ; Figure 2C). The scoring of risk-taking behavior during the second stage of the experiment indicated that, during the 30 min after the scaring stimuli, the distance covered by crayfish ranged from 0 cm to 760 cm in the neutral context, while it ranged from 0 cm to 1505 cm in the food context. Risk-taking behavior was not significantly repeatable across contexts ( $R = 0.06$  [0–0.40];  $P = 0.375$ ;  $n = 24$ ; Figure 2D).

Relative early nocturnal activity did not show significant correlations with risk-taking behavior in either the neutral ( $r_\tau = -0.12$ ;  $P = 0.398$ ; Figure 3) or food context ( $r_\tau = 0.19$ ;  $P = 0.205$ ; Figure 3). Similarly, relative middle nocturnal activity did not show significant correlations with risk-taking behavior in either the neutral ( $r_\tau = -0.01$ ;  $P = 0.960$ ; Figure 3) or food context ( $r_\tau = 0.02$ ;  $P = 0.862$ ; Figure 3). In contrast, relative late nocturnal activity showed a significant positive correlation with risk-taking behavior in the food context ( $r_\tau = 0.34$ ;  $P < 0.05$ ; effect size  $A = 0.125$ ; Figure 3) while it did not show significant correlations with risk-taking behavior in the neutral context ( $r_\tau = 0.15$ ;  $P = 0.296$ ; Figure 3). It is important to note that relative nocturnal activity was not correlated with the mean overall activity (early:  $r_\tau = -0.04$ ;  $P = 0.766$ ; middle:  $r_\tau = -0.01$ ;  $P = 0.921$ ; late:  $r_\tau = 0.25$ ;  $P = 0.082$ ), which means that the chronotypes described here are not related to activity phenotypes. Furthermore, the mean overall activity and the mean nocturnal activity were not correlated with risk-taking behavior either in the neutral (mean overall activity:  $r_\tau = 0.15$ ;  $P = 0.320$ ; mean nocturnal activity:  $r_\tau = 0.07$ ;  $P = 0.654$ ) or food context (mean overall activity:  $r_\tau = 0.19$ ;  $P = 0.205$ ; mean nocturnal activity:  $r_\tau = -0.11$ ;  $P = 0.441$ ).

## Discussion

We showed that daily activity rhythms can have consistent inter-individual differences in terms of relative amount of locomotor activity across days as indicative of chronotypes (Helm et al. 2017). However, our results indicate some differences related to the specific activity period. In particular, we showed a significant repeatability for relative early and middle nocturnal activity, but not for relative late nocturnal activity, suggesting that crayfish being unpredictable in the last part of the night could have some ecological advantage related to predation (Briffa 2013). Moreover, we did not find consistent inter-individual differences in risk-taking behavior across contexts, suggesting that crayfish behavior expressed in the two contexts could have different ecological significance; therefore, we cannot consider risk-taking behavior as a personality trait.



**Figure 1.** Daily activity of rhythmic (A) and arrhythmic (B) crayfish together with 3 representative examples of daily activity rhythms: (C) ID12, which showed the highest relative early nocturnal activity; (D) ID6, which showed the highest relative middle nocturnal activity; (E) ID19, which showed the second highest relative late nocturnal activity. Average locomotor activity (points) is shown together with standard errors (vertical bars) and the midline estimating statistic of rhythm (i.e., mesor, the central tendency of locomotor activity; horizontal line). The gray areas represent darkness hours. The vertical dashed lines represent the time periods used to calculate relative nocturnal activity. The arrows (C–E) represent the period of darkness where the crayfish was more active compared to the other crayfish.

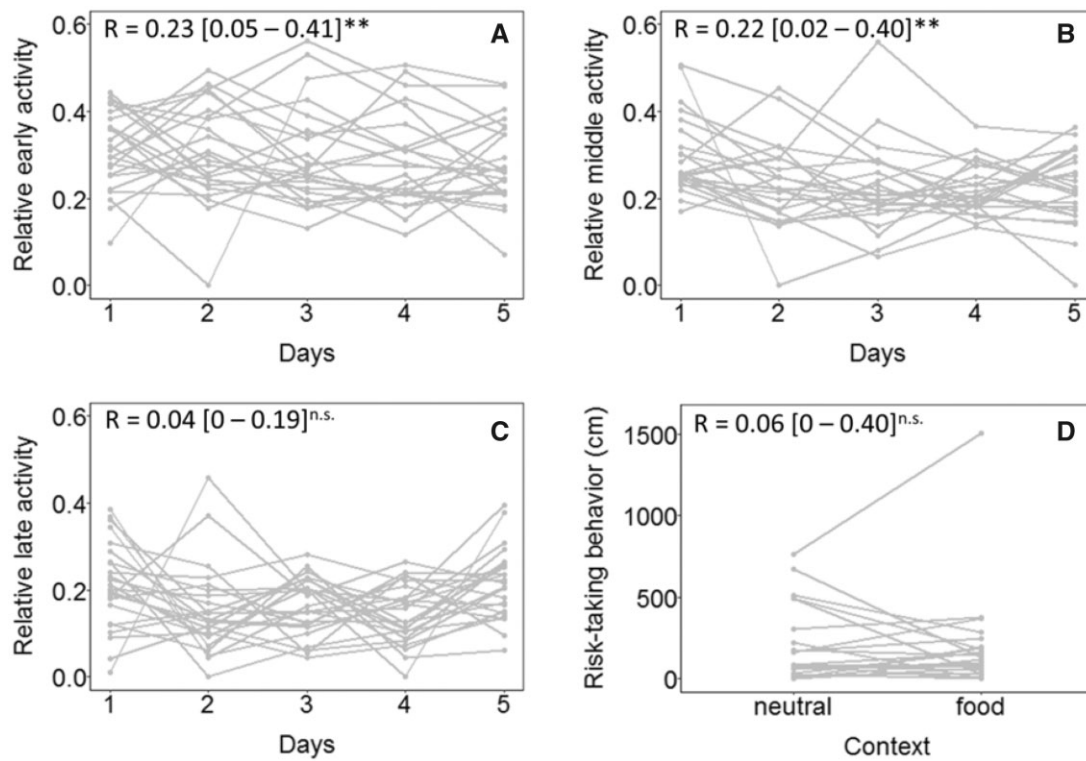
Consequently, we cannot formally assess the presence of a behavioral syndrome between chronotypes and risk-taking behavior. However, we showed that relative late nocturnal activity was positively correlated with risk-taking behavior, but only in the presence of food.

We provide evidence that the relative amount of locomotor activity displayed in specific activity period can also show consistent inter-individual differences. The importance of consistent inter-individual differences in the timing of activity has recently been highlighted (Helm et al. 2017). Our results agree with previous evidence on Zebrafish where consistency of locomotor activity rhythms has been tested at the group level (Sbragaglia et al. 2021). Interestingly, we showed that the consistent inter-individual differences in early and middle nocturnal activity accounted for 23 and 22% of the phenotypic variance across time, respectively. The degree of consistency corresponds to personality traits of other species in a wide range of animal taxa (Bell et al. 2009). It must be noted that the chronotypes measured here do not correlate to the mean level of overall activity (i.e., midline estimating statistic of rhythm). Therefore, it represents a phenotype that is not related to inter-individual differences of overall activity, but to inter-individual differences in the allocation of activity throughout the day.

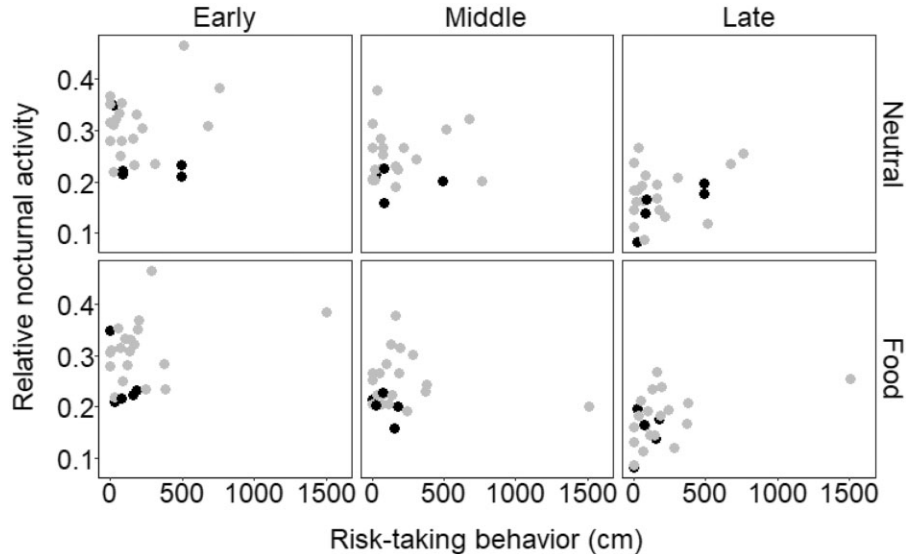
Our results suggest that consistent inter-individual differences in the relative amount of locomotor activity allocated to different time

of the day could be an important factor in determining the ecological significance of biological rhythms. Indeed, biological rhythms research mainly focused on the high degree of plasticity of daily locomotor activity rhythms. For example, temporal niche switching (i.e. shifts in the timing of daily locomotor activity) has been suggested as a strategy to maximize survival in response to environmental changes both in endotherms and ectotherms (Chiesa et al. 2010; Hut et al. 2012; van der Vinne et al. 2019). However, the consistency in the relative amount of locomotor activity allocated to different time of the day has been largely overlooked, limiting the potential to understand ecological significance of biological rhythms. Indeed, behavioral ecologists have already recognized that both consistent inter-individual differences (i.e., personality) and individual plasticity must be considered as complementary aspects of individual phenotypes (Dingemans et al. 2010). More research aiming in disentangling the relative proportion of consistency and plasticity of daily activity rhythms is needed to fully understand their ecological significance.

Our results do not show consistency of behavior across contexts and therefore we cannot consider risk-taking behavior as a personality trait. A possible explanation is that what we measure are two different behaviors; for example, the amount of locomotor activity performed by crayfish after the scaring stimulus could be related to exploration or activity per se. In contrast, in the presence of food,



**Figure 2.** Repeatability of relative early (A), middle (B), and late (C) nocturnal activity across 5 days ( $n = 24$ ) together with the repeatability of risk-taking behavior across contexts (D;  $n = 24$ ). Each line represents an individual crayfish together with the repeatability score, confidence interval, and significance level (\*\* $P < 0.01$ ; ns: not significant).



**Figure 3** Scatter plot of relative nocturnal activity (early, middle, late) and risk-taking behavior measured in 2 different contexts (neutral and food). Gray points represent the crayfish with rhythmic daily activity rhythms ( $n = 19$ ; see also Figure 1A), while black points represent crayfish with arrhythmic daily activity rhythms ( $n = 5$ ; see also Figure 1B).

locomotor activity could be related to feeding behavior. Therefore, we were not able to formally test the existence of a behavioral syndrome between chronotypes and risk-taking behavior (Dingemans and Wright 2020). However, we have found that crayfish that were overall more active at the end of darkness were also more risk-takers in the presence of food. This may suggest that activity late in the darkness is somehow related to risk-taking behavior. Such

interpretation is also reinforced by the fact that crayfish were not repeatable in their relative late nocturnal activity across days. Indeed, Briffa (2013) suggested that unpredictable individual behavior might represent a strategy for dealing with risk. Therefore, being unpredictable late in the night and link the amount of locomotor activity to risk-taking behavior—only in the presence of food—could be an ecological strategy for the signal crayfish to cope with predators.

Indeed, some of the major predators of adult crayfish are raccoons, otters, and eels (Jones et al. 2016; Jurcak et al. 2016), which rely on mechanical and chemical senses to hunt at night. Important anti-predatory behaviors of crayfish include reduced movement or seeking cover (Breithaupt et al. 2016; Jurcak et al. 2016). Moreover, crayfish exposed to fish predators spent less time out of the refuge at night than individuals not exposed to predators (Nyström 2005). Therefore, it is plausible assuming that the locomotor activity recorded out of the refuge in our experiments may be linked to risk-taking behavior of this species. Indeed, the signal crayfish responded to the scaring stimulus by reducing locomotor activity or by searching cover into the shelter. Our interpretation is purely speculative and such topic deserves more research attention in the future.

There are two limitations in our experimental approach that need to be highlighted. The first one is related to the fighting history of the individuals during the acclimation period. Indeed, the crayfish used in the experiments were co-housed in the same acclimation facility before running the individual behavioral trials. Although they had individual refuges and enough food, we observed conspecific fights. Consequently, we cannot exclude that established dominance hierarchies affected individual behavior recorded after the acclimation period. Although dominance hierarchies seem to not have effects on daily rhythms of emergence from refuges in lobsters (Sbragaglia et al. 2017), we cannot exclude that fighting history had an effect on risk-taking behavior. The second limitation is related to the weak statistical support for the correlation between late nocturnal and risk-taking behavior in the food context. Research is needed to provide more robust support for such link between daily activity rhythms and risk-taking behavior.

In conclusion, we provided the first evidence of chronotypes associated to the relative amount of activity displayed in specific time of the day. Moreover, while the signal crayfish was already known to be a nocturnal species (Nyström 2005; Edmonds et al. 2011; Thomas et al. 2016), here we provided an in-depth quantitative measure of daily activity rhythms. Animal behavior plays a paramount role in biological invasions (Holway and Suarez 1999; Linzmaier et al. 2018; Ruland and Jeschke 2020), and personality traits can determine the success or failure at different stages of biological invasions (Chapple et al. 2012; Carere and Gherardi 2013; Jutte et al. 2015; Canestrelli et al. 2016). In contrast, the role of daily activity rhythms in biological invasions is unknown. Considering that the signal crayfish is a major invasive species, the daily activity patterns and chronotypes documented here could be useful to understand the behavioral mechanisms behind the invasiveness of this species.

## Ethical Statement

Experiments were conducted at the University of Hull and approved by the Ethics committee of the Department of Biological Sciences (approval number U035).

## Data Accessibility

Data are available as [Supplementary Material](#) together with the R code.

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## Authors' Contribution

T.B. and V.S. conceived the idea; V.S. ran the experiments, analyzed the data, and wrote the manuscript with inputs of T.B.

## Competing Interest

We declare no competing interests.

## References

- Bell AM, Hankison SJ, Laskowski KL, 2009. The repeatability of behaviour: a meta-analysis. *Anim Behav* 77:771–783.
- Breithaupt T, Gherardi F, Aquiloni L, Tricarico E, 2016. Chemical ecology of crayfish. In: Longshaw M, Stebbing P, editors. *Biology and Ecology of Crayfish*. Boca Raton (FL): CRC Press. 132–170.
- Briffa M, 2013. Plastic proteans: reduced predictability in the face of predation risk in hermit crabs. *Biol Lett* 9:20130592.
- Briffa M, Rundle SD, Fryer A, 2008. Comparing the strength of behavioural plasticity and consistency across situations: animal personalities in the hermit crab *Pagurus bernhardus*. *Proc R Soc Lond B* 275:1305–1311.
- Canestrelli D, Bisconti R, Carere C, 2016. Bolder takes all? The behavioral dimension of biogeography. *Trends Ecol Evol* 31:35–43.
- Carere C, Gherardi F, 2013. Animal personalities matter for biological invasions. *Trends Ecol Evol* 28:5–6.
- Castillo-Ruiz A, Paul MJ, Schwartz WJ, 2012. In search of a temporal niche: social interactions. *Prog Brain Res* 199:267–280.
- Chapple DG, Simmonds SM, Wong BBM, 2012. Can behavioral and personality traits influence the success of unintentional species introductions? *Trends Ecol Evol* 27:57–64.
- Chiesa JJ, Aguzzi J, García JA, Sardà F, de la Iglesia HO, 2010. Light intensity determines temporal niche switching of behavioral activity in deep-water *Nephrops norvegicus* (crustacea: decapoda). *J Biol Rhythms* 25:277–287.
- Dingemanse NJ, Kazem AJN, Réale D, Wright J, 2010. Behavioural reaction norms: animal personality meets individual plasticity. *Trends Ecol Evol* 25: 81–89.
- Dingemanse NJ, Wright J, 2020. Criteria for acceptable studies of animal personality and behavioural syndromes. *Ethol*. 126:865–869.
- Dunlap JC, Loros JJ, DeCoursey PJ, 2004. *Chronobiology: Biological Timekeeping*. Sunderland (MA): Sinauer Associates.
- Edmonds N, Riley W, Maxwell D, 2011. Predation by *Pacifastacus leniusculus* on the intra-gravel embryos and emerging fry of *Salmo salar*. *Fish Manage Ecol* 18:521–524.
- Fero K, Simon JL, Jourdie V, Moore PA, 2007. Consequences of social dominance on crayfish resource use. *Behaviour* 144:61–82.
- Helm B, Visser ME, Schwartz W, Kronfeld-Schor N, Gerkema M et al., 2017. Two sides of a coin: ecological and chronobiological perspectives of timing in the wild. *Phil Trans R Soc B* 372:20160246.
- Holdich DM, 2002. *Biology of Freshwater Crayfish*. Oxford: Blackwell Science.
- Holway DA, Suarez AV, 1999. Animal behavior: an essential component of invasion biology. *Trends Ecol Evol* 14:328–330.

- Hut RA, Kronfeld-Schor N, van der Vinne V, De la Iglesia H, 2012. In search of a temporal niche: environmental factors. *Prog Brain Res* 199:281–304.
- Jones EW, Jackson MC, Grey J, 2016. Environmental drivers for population success: population biology, population and community dynamics. In: Longshaw M, Stebbing P, editors. *Biology and Ecology of Crayfish*. CRC Press, London. 251–286.
- Juette T, Cucherousset J, Cote J, 2015. Animal personality and the ecological impacts of freshwater non-native species. *Curr Zool* 60:417–427.
- Jurcak AM, Lahman SeWofford SJ, Moore PA, 2016. Behavior of crayfish. In: Longshaw M, Stebbing B, editors. *Biology and Ecology of Crayfish*. Boca Raton: CRC Press. 117–131.
- Kronfeld-Schor N, Bloch G, Schwartz WJ, 2013. Animal clocks: when science meets nature. *Proc R Soc Lond B* 280:20131354.
- Kronfeld-Schor N, Dayan T, 2003. Partitioning of time as an ecological resource. *Annu Rev Ecol Evol Syst* 34:153–181.
- Levy O, Dayan T, Rotics S, Kronfeld-Schor N, 2012. Foraging sequence, energy intake and torpor: an individual-based field study of energy balancing in desert golden spiny mice. *Ecol Lett* 15: 1240–1248.
- Linzmaier SM, Goebel LS, Ruland F, Jeschke JM, 2018. Behavioral differences in an over-invasion scenario: marbled vs. Spiny-cheek crayfish. *Ecosphere* 9: e02385.
- Martorell-Barceló M, Campos-Candela A, Alós J, 2018. Fitness consequences of fish circadian behavioural variation in exploited marine environments. *PeerJ* 6:e4814.
- Moiran M, Laskowski KL, Niemela PT, 2020. Individual differences in behaviour explain variation in survival: a meta-analysis. *Ecology Letters* 23: 399–408.
- Mowles SL, Cotton PA, Briffa M, 2012. Consistent crustaceans: the identification of stable behavioural syndromes in hermit crabs. *Behav Ecol Sociobiol* 66:1087–1094.
- Nakagawa S, Schielzeth H, 2010. Repeatability for Gaussian and non-Gaussian data: a practical guide for biologists. *Biol Rev Camb Philos Soc* 85:935–956.
- Nyström P, 2005. Non-lethal predator effects on the performance of a native and an exotic crayfish species. *Freshwat Biol* 50:1938–1949.
- Pintor LM, Sih A, Kerby JL, 2009. Behavioral correlations provide a mechanism for explaining high invader densities and increased impacts on native prey. *Ecology* 90:581–587.
- Réale D, Garant D, Humphries MM, Bergeron P, Careau V et al., 2010. Personality and the emergence of the pace-of-life syndrome concept at the population level [10.1098. 0208]. *Philos Trans R Soc Lond B Biol Sci* 365: 4051–4063.rstb.2010.
- Refinetti R, 2006. *Circadian Physiology*. 2nd edn. Boca Raton (FL): CRC Press.
- Ruland F, Jeschke JM, 2020. How biological invasions affect animal behaviour: a global, cross-taxonomic analysis. *J Anim Ecol* 89:2531–2541.
- Sbragaglia V, Aguzzi J, García J, Sarriá D, Gomariz S et al., 2013. An automated multi-flume actograph for the study of behavioral rhythms of burrowing organisms. *J Exp Mar Biol Ecol* 446:177–185.
- Sbragaglia V, García J, Chiesa J, Aguzzi J, 2015a. Effect of simulated tidal currents on the burrow emergence rhythms of the norway lobster *Nephrops norvegicus*. *Mar Biol* 162:2007–2016.
- Sbragaglia V, Lamanna FM, Mat A, Rotllant G, Joly S et al., 2015b. Identification, characterization, and diel pattern of expression of canonical clock genes in *Nephrops norvegicus* (crustacea: decapoda) eyestalk. *PLoS ONE* 10:e0141893.
- Sbragaglia V, Leiva D, Arias A, Garcia JA, Aguzzi J et al., 2017. Fighting over burrows: the emergence of dominance hierarchies in the norway lobster *Nephrops norvegicus*. *J Exp Biol* 220:4624–4633.
- Sbragaglia V, Lopez-Olmeda JF, Frigato E, Bertolucci C, Arlinghaus R, 2021. Size-selective mortality induces evolutionary changes in group risk-taking behaviour and the circadian system in a fish. *J Anim Ecol* 90:387–403.
- Smith BR, Blumstein DT, 2008. Fitness consequences of personality: a meta-analysis. *Behavioral Ecology* 19:448–455.
- Sokolove PG, Bushell WN, 1978. The chi square periodogram: its utility for analysis of circadian rhythms. *J Theor Biol* 72:131–160.
- Stein RA, Magnuson JJ, 1976. Behavioral response of crayfish to a fish predator. *Ecology* 57:751–761.
- Thomas JR, James J, Newman RC, Riley WD, Griffiths SW et al., 2016. The impact of streetlights on an aquatic invasive species: artificial light at night alters signal crayfish behaviour. *Appl Anim Behav Sci* 176:143–149.
- Tudorache C, Slabbekoorn H, Robbers Y, Hin E, Meijer JH et al., 2018. Biological clock function is linked to proactive and reactive personality types. *BMC Biol* 16:148.
- Vainikka A, Rantala MJ, Niemelä P, Hirvonen H, Kortet R, 2011. Boldness as a consistent personality trait in the noble crayfish, *astacus astacus*. *Acta Ethologica* 14:17–25.
- van der Vinne V, Tachinardi P, Riede SJ, Akkerman J, Scheepe J et al., 2019. Maximising survival by shifting the daily timing of activity. *Ecol Lett* 22: 2097–2102.
- Vargha A, Delaney HD, 2000. A critique and improvement of the cl common language effect size statistics of mcgraw and wong. *J Educ. Behav Stat* 25: 101–132.

