

Fixed vs. Random Temporal Predictability of Predation Risk: An Extension of the Risk Allocation Hypothesis

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Abstract

Predation is a strong selective force acting on prey animals. Predation is by nature highly variable in time; however, this aspect of predation risk has traditionally been overlooked by behavioural ecologists. Lima and Bednekoff proposed the predation risk allocation hypothesis (RAH), predicting how temporal variation in predation risk drives prey antipredator behaviours. This model is based on the concept that prey adaptively allocate their foraging and antipredator efforts across high- and low-risk situations, depending on the duration of high- vs. low-risk situations and the relative risk associated with each of them. An unstudied extension of the RAH is the effect of predictability of predation risk. A predictable risk should lead to prey displaying minimal vigilance behaviours during predictable low-risk periods and the strongest antipredator behaviours during risky periods. Conversely, an unpredictable predation risk should result in prey displaying constant vigilance behaviour, with suboptimal foraging rates during periods of safety but antipredator behaviours of lower intensity during periods of risk. We tested this extension of the RAH using convict cichlids exposed to high-risk alarm cues at two frequencies of risk (1× vs. 3×) per day, on either a fixed or random schedule for 5 d. We then tested the fish for a response to high-risk cues (alarm cues) and to low-risk cues (disturbance resulting from the introduction of distilled water). Our study supports previous results on the effects of risk frequency and cue intensity on cichlid behaviour. We failed to show an effect of risk predictability on the behavioural responses of cichlids to high-risk alarm cues, but predictability did influence responses to low-risk cues. We encourage further studies to test the effect of predictability in other systems.

Introduction

Predation is a major evolutionary force acting on prey behaviour (Lima & Dill 1990). Behavioural decisions made under the risk of predation have been a major topic of interest in behavioural ecology and many studies have attempted to understand how prey animals balance necessary but risky activities, such as feeding and mating, with predator

avoidance. Most of these studies have investigated the change in the prey's behaviours in the presence vs. absence of predation risk (Chivers & Smith 1998). However, predation is by nature variable in space and time. Predators come and go on a moment-to-moment basis or on a diurnal, lunar or seasonal cycle, and this aspect of predation risk has been widely overlooked by behavioural ecologists (Sih et al. 2000). Until recently, no model has

focused explicitly on the effect of temporal variation in predation risk on prey behaviours. Such a model would have to get away from the classical static study of predator–prey relationships and incorporate the dynamic temporal nature of such interactions.

In 1999, Lima and Bednekoff proposed the predation risk allocation hypothesis (RAH), explaining how temporal variation in predation risk drives prey antipredator behaviours. This model is based on the concept that prey adaptively allocate their foraging efforts and therefore, their exposure to predation risk, across high- and low-risk situations. Prey behaviours in any situation should depend on the overall risk experienced by prey, more specifically the duration of high- vs. low-risk situations and the relative risk associated with each of them. The RAH predicts that as the duration of exposure to a risk increases, prey should decrease their antipredator response intensity, as long periods of antipredator behaviour may result in unacceptable decreases in foraging activities. Moreover, animals exposed to long periods of high risk should devote more foraging activity during brief periods of safety, compared with prey exposed to infrequent risk. The model also predicts that, as the relative risk associated with the high-risk situation increases compared with the low-risk situation, prey should increase their antipredator response intensity, but will consequently increase their foraging effort in the low-risk situation to compensate for the loss of foraging opportunities.

Following its publication, a number of studies attempted to empirically test Lima and Bednekoff's model. Studies have encompassed a diversity of organisms, including both invertebrate (e.g. Hamilton & Heithaus 2001; Sih & McCarthy 2002; Pecor & Hazlett 2003) and vertebrate (e.g. Van Burskirk et al. 2001; Koivisto & Pusenius 2003, Sundell et al. 2004) taxa. The results of these studies often show only partial support for the RAH model (Mirza et al. 2006; Pecor & Hazlett 2006; Slos & Stoks 2006). In one study, Foam et al. (2005) found support for the RAH testing convict cichlids (*Archocentrus nigrofasciatus*). In their study, they exposed cichlids to two frequencies of risk (once a day and three times a day) and two levels of risk (100% and 20% concentration of a conspecific alarm cue solution). They found that, during risky periods, fish exposed to predation risk three times a day exhibited lower antipredator responses than those exposed to predation risk once a day when exposed to the 100% alarm cue solution. Moreover, they found that, during periods of safety, fish exposed to risk three times a day foraged more than those exposed to risk only once a day.

Thus, it seems that the cichlid/alarm cue prey/risk model is a good fish system to further test the RAH.

One aspect of the RAH that has not been considered is how temporal predictability of the threat affects the behavioural responses of prey individuals. A number of predators are known to actively forage during specific periods of the day, e.g. during dawn and dusk only or alternatively during daytime or night-time. Prey have also been known to respond to temporally variable risk of predation by altering their habitat use, foraging habits, reproduction and territorial defence (Lima & Dill 1990). A classic example is copepods exhibiting diel vertical migration as an antipredator response to escape from predators (Neill 1990). From an RAH perspective, if a predation threat is temporally predictable, i.e. the prey knows when the predator will appear, then the prey should display a minimal or absent vigilance behaviour and a maximum foraging behaviour prior to the predator appearance, and display high-intensity antipredator behaviour during the threat period. On the other hand, if the predation threat is unpredictable, prey might always display vigilance behaviour and a suboptimal foraging behaviour. This would result in prey displaying a lower intensity response to the predator compared with those with a predictable predator.

In this study, we attempted to test the predator predictability aspect of the RAH by exposing prey to either a temporally fixed or random predation risk for five consecutive days. As we did not know whether frequency of risk or the intensity of the risk would interact with the predictability of risk, to influence the behaviour of the prey, we decided to incorporate these variables in our experiment. Foam et al. (2005) established that the frequency of risk and the intensity of risk influence cichlid behaviour as predicted by the RAH. Hence, we used the same system in our experiment, exposing convict cichlids to two frequencies of risk (once or three times a day) and to two predictabilities of risk (fixed or random). We then tested each of these four groups of fish to high-risk cues (alarm cues from conspecifics) and low-risk cues (distilled water, which represents a generalized disturbance cue).

Methods

Test Fish and Stimulus Preparation

We used juvenile convict cichlids which were descendants of laboratory stock populations crossed with wild cichlids originally from Costa Rica.

The cichlids were held in 110-l aquaria containing continuously filtered dechlorinated tap water (24–26°C, pH 7.1–7.3) and a gravel substrate. Fish in holding tanks were exposed to a 12-h:12-h L:D cycle and were fed *ad libitum* twice a day with commercial flake food and brine shrimp (*Artemia* spp.).

We collected chemical alarm cue from the skin of 34 donor cichlids (mean \pm SD standard length = 4.73 \pm 0.47 cm). The collecting procedure was the same as used in Foam et al. (2005) and Brown et al. (2006a,b). We collected a total of 247.7 cm² of skin and adjusted the final volume to 2810 ml with distilled water. Alarm cue was frozen in 20-ml aliquots at –20°C until needed.

Creating Background Levels of Risk

Conditioning tanks consisted of a series of white opaque plastic basins (48 \times 38 \times 20 cm³), filled with 23 l of dechlorinated tap water. Conditions were the same as described above, but the conditioning tanks were not filtered. Each basin was equipped with an overflow valve, positioned approximately 5 cm from the upper edge, to facilitate water changes.

To create different levels of risk, we exposed groups of 25 cichlids to high-risk conspecific alarm cue at either a low frequency (once per day) or high frequency (three times per day). In addition, for both levels of frequency, we gave the cue in either a fixed or random schedule. For the ‘fixed’ treatment, cichlids were exposed to the alarm cue at 12:00 hours (low frequency) or at 09:00, 12:00 and 15:00 hours daily (high frequency), for five consecutive days. For the ‘random’ treatment, we determined the time of exposure using a random number generator. Exposures (once or three times a day according to the frequency treatment) were made at least 1 h apart, between 08:00 and 16:00 hours daily, with the proviso that the temporal pattern could not be identical over two consecutive days. Thus, we created four levels of background risk: (1) fixed low frequency (1F); (2) random low frequency (1R); (3) fixed high frequency (3F); and (4) random high frequency (3R). Although only 20 cichlids from each conditioning basin would be tested, we conditioned a total of 12 groups of 25 to ensure that we could closely match test fish for size (see below). Each conditioning ‘group’ of 20 test fish yielded five pairs of cichlids tested to the high-risk alarm cue treatment and five pairs tested to the low-risk distilled water control treatment.

The alarm cue stimulus was injected next to the airstone in a series of three pulses, each separated by

10 min. We gave the injections as a series of three 10-ml injections of alarm cue to better simulate natural patterns of risk (Foam et al. 2005). Ten minutes after each series of injections, we slowly flushed the basins with approximately 25 l of dechlorinated water. Water changes were carried out on all conditioning tanks, regardless of the treatment to control for any disturbance effects. During this pre-treatment phase, cichlids were fed twice daily with commercial flake food. Food was never given in combination with the alarm cues. Approximately 1 h after the final exposure on the fifth day, we transferred pairs of cichlids, matched for size, to test tanks and allowed them to acclimate overnight before testing on the sixth day. Mean (\pm SD) standard length at testing was 3.41 \pm 0.37 cm.

Testing Phase

Fish were tested on day 6 at 12:00 hours. Test tanks consisted of a series of 37-l glass aquaria, equipped with a single airstone, a gravel substrate and an additional length of tubing to allow for the injection of chemical stimuli from a distance of at least 2 m. Test tanks were filled with dechlorinated tap water, but were not filtered. Temperature and lighting were identical to the holding tanks. We divided each tank into three vertical sections with lines drawn on the exterior of the tanks to facilitate recording area use (see below). In addition, we covered three sides of each test tank with opaque plastic to ensure visual isolation between test tanks. Fish were fed equal amounts of commercial flake food the evening before the test day, as well as 1 h prior to testing. Test fish were fed enough flake food to ensure that some flakes, once saturated with water, would sink to the substrate providing foraging opportunities during the trials. We conducted between 11 and 15 replicates per treatment, and individual cichlids were tested only once.

Trials consisted of a 5-min pre-stimulus observation period, followed by a 5-min post-stimulus observation period (see Brown et al. 2006a,b for further details). Following the pre-stimulus observation period, we injected 10 ml of distilled water (low-risk cue, representing a disturbance effect) or 10 ml of conspecific alarm cue (high-risk treatment). During both the pre- and post-stimulus injection observation periods, we recorded the number of foraging attempts (as the per capita rate per minute), vertical area use and time spent moving. We defined a foraging attempt as the pecking at the gravel substrate, with the body inclined at an angle greater than 45°

to the substrate (Grant et al. 2002). Area use was recorded every 15 s by assigning each fish a score of 1 (bottom third of the tank) to 3 (top third of the tank). As such, mean area use scores ranged from 2 (both fish near the substrate) to 6 (both fish near the surface). We recorded time moving as the mean time (in seconds) either of the pair of test cichlids spent not being stationary (Brown et al. 2006a,b). Decreased time moving and frequency of foraging attempts as well as increased time spent near the bottom are indicative of antipredator responses in juvenile cichlids (Wisenden & Sargent 1997; Alemadi & Wisenden 2002; Brown et al. 2004).

Statistical Analysis

The data for all three behavioural measurements were normally distributed. Data for foraging attempts and time moving had homogeneous variances among treatments. Moreover, the data for these two variables are likely to be highly correlated; thus, we conducted a two-way multivariate analysis of variance for the analyses of these data, to investigate the effect of predictability and frequency on the responses of cichlids to the high- and low-risk cues. Data for area use did not have homogeneous variances; thus, we performed a two-way Scheirer–Ray–Hare extension of the Kruskal–Wallis test (Sokal & Rohlf 2003), which is a multiway ANOVA design for ranked data. We analysed the data for water and alarm cues separately, as we found a three-way interaction.

Results

The results of the multivariate ANOVA on foraging attempts and time moving of cichlids exposed to the low-risk cues (water disturbance) showed a significant effect of predictability ($F_{2,43} = 3.4$, $p = 0.043$, Figs 1 and 2), but no significant effect of frequency ($F_{2,43} = 0.3$, $p = 0.770$) and no interaction between the two factors ($F_{2,43} = 1.6$, $p = 0.206$). The effect of predictability comes from the fact that cichlids increased time moving more when the injection of water was fixed, compared with the random injection ($F = 4.6$, $p = 0.037$). However, predictability did not have an effect on foraging attempts ($F = 0.6$, $p = 0.439$).

The results of the multivariate ANOVA on foraging attempts and time moving of cichlids exposed to high-risk alarm cues showed no significant effect of predictability ($F_{2,48} = 0.4$, $p = 0.670$), a significant effect of frequency ($F_{2,48} = 6.0$, $p = 0.005$) but no interaction between the two factors ($F_{2,48} = 0.7$,

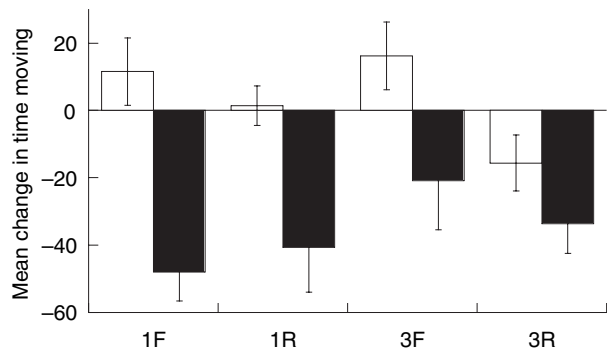


Fig. 1: Mean (\pm SE) change in time spent moving (from the pre-stimulus baseline) for cichlid pairs responding to either distilled water (empty bars) or alarm cues (solid bars). Prior to testing, cichlids were exposed for 5 d to a predation risk once a day (1) or three times a day (3) on either a fixed (F) or random (R) schedule.

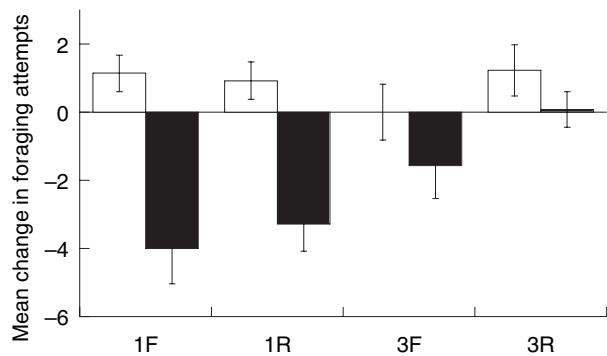


Fig. 2: Mean (\pm SE) change in the number of foraging attempts (from the pre-stimulus baseline) for cichlid pairs responding to either distilled water (empty bars) or alarm cues (solid bars). Prior to testing, cichlids were exposed for 5 d to a predation risk once a day (1) or three times a day (3) on either a fixed (F) or random (R) schedule.

$p = 0.509$). Cichlids exposed to alarm cues in the high-frequency treatments responded with a lower intensity of antipredator response than the cichlids in the low-frequency treatments.

The results of the Scheirer–Ray–Hare test performed on the area use data revealed no effect of frequency ($F_{1,53} = 2.6$, $p = 0.110$; $F_{1,53} = 1.6$, $p = 0.213$, Fig. 3), no effect of predictability ($F_{1,53} = 0.3$, $p = 0.612$; $F_{1,53} = 0.0$, $p = 0.980$) and no interaction between the two factors ($F_{1,53} = 0.0$, $p = 0.980$; $F_{1,53} = 0.3$, $p = 0.565$) when cichlids were exposed to water or alarm cues respectively.

Discussion

The results of our study show that frequency of risk (i.e. exposure to alarm cues) was an important

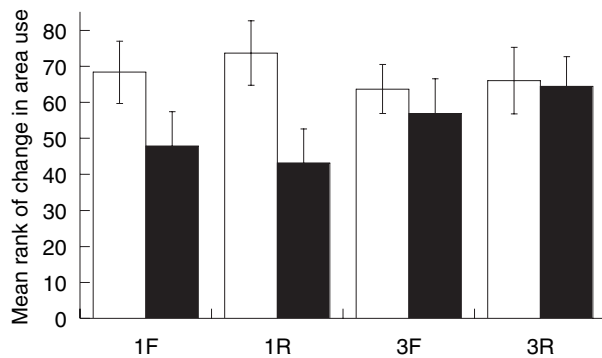


Fig. 3: Mean (\pm SE) rank of the change in area use (from the pre-stimulus baseline) for cichlid pairs responding to either distilled water (empty bars) or alarm cues (solid bars). Prior to testing, cichlids were exposed for 5 d to a predation risk once a day (1) or three times a day (3) on either a fixed (F) or random (R) schedule.

variable influencing the behaviour of cichlids. Our results are consistent with those of Foam et al. (2005) and Brown et al. (2006a,b). Indeed, we found that cichlids exposed to the high-frequency treatments responded with a lower intensity of anti-predator response in terms of area use than those in the low-frequency treatments. Moreover, when given predation cues, cichlids in the low-frequency treatments reduced their foraging effort more than cichlids in the high-frequency treatments. These responses are consistent with the prediction of the RAH (Lima & Bednekoff 1999).

In our experiment, we did not find any effect of risk predictability on any of the response variables when cichlids were exposed to high-risk alarm cues. However, there was an effect of predictability on the responses of cichlids to distilled water. Cichlids increased time moving more when the injection of water was fixed compared with the random injection. The results highlight that the effects of predictability are influenced by the intensity of the risk cue the fish are exposed to. The absence of a predictability effect to the high-risk predation cues could be the result of a relatively short conditioning period. Indeed, 5 d may be a short time for cichlids to accurately gauge predictability, although we did observe a predictability effect when cichlids were exposed to low-risk water. Moreover, this length of time was clearly long enough for cichlids to gauge frequency. We suggest that future researchers should consider predictability and how it interacts with risk intensity, but should ensure a long enough acclimation time for their test species. In our experiment, all of the risky periods occurred during the day; hence, our ability to identify a predictability effect may have

been weakened by the limited time frame (8 h) used in our experiment. It might be more relevant to provide predictability cues at different times during the 24-h cycle. For example, fish may cue more easily on predation risk provided at dawn and dusk than at different times within the light period. Future researchers need to carefully consider the time frame they choose for examining predictability is appropriate for their test species.

The premise of our experiment begs the question about whether predation risk is indeed predictable. This topic has received relatively little attention. We have good evidence that some predators forage on a specific schedule (Lima & Dill 1990). For example, some piscivores are specialized crepuscular predators (feed at dawn and dusk). Others are specialized nocturnal or diurnal predators. Prey exposed solely to a specific kind of predator might benefit from the predictability of foraging patterns of those predators. However, a specific ecosystem might contain a wide array of predators. Even though some may be crepuscular, other predators occurring in the same habitat may be diurnal or nocturnal. Hence, it might be somewhat difficult for prey to accurately gauge predictability. The behaviour of convict cichlids has been studied extensively in the laboratory. However, relatively few studies have been conducted in the field. While these studies have considered predation (Wisenden & Keenleyside 1994), we have no information about the temporal predictability of predation this species normally encounters.

In our experiment, we provided predation risk cues in the form of disturbance from injection of water to the tank and from alarm cues from injured conspecifics. Alarm cues provide a generalized indication of predation risk (reviewed by Chivers & Smith 1998; Wisenden & Chivers 2006). However, they do not provide the prey with information on the identity of the predator that caused the release of the cues. This may confound the prey's ability to assess predictability. Future studies should examine predictability with cues of specific predators, particularly those that co-occur with the prey and have a well-defined foraging pattern.

Chemicals provide a great source of information for animals living in aquatic environments (Chivers & Smith 1998; Wisenden & Chivers 2006). Chemical information persists longer in the medium than visual or electrical information and, contrary to visual cues, is available day and night, in all types of habitats. However, chemical information is probably less reliable spatially and temporally than other cues (Ferrari et al. in press). By their very nature,

chemicals cues might not allow fish to accurately assess predictability in a reliable fashion. We suggest that future research should attempt to use visual cues to test the effects of predictability.

The RAH provides a powerful framework for understanding how temporal variability in predation risk influences foraging and antipredator responses of prey. We found an effect of predictability using low-risk cues but not high-risk cues. This research highlights the need for other researchers to consider predictability in their studies of risk allocation.

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