Social learning about predators: does timing matter?

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In Pavlovian conditioning, animals acquire a response to a previously neutral stimulus (conditioned stimulus, CS), such as a light, if that stimulus predicts a biologically important event (unconditioned stimulus, US), such as delivery of food. Learning typically occurs when the CS precedes the US (forward conditioning), and not when the CS follows the US (backward conditioning). In social learning about predators, the predator stimulus is considered to be the CS to which observers acquire avoidance responses after the stimulus has been presented in contiguity with an alarmed demonstrator, the US. We tested the prediction that social learning of response to a predator would occur even if the social alarm cues (the US) appeared before the predatory stimulus (the CS). Carib grackles, *Quiscalus lugubris*, responded to a familiar predator presented at close range by suppressing alarm calls. Presentation of an unfamiliar avian model (black-and-yellow pigeon) also decreased calling, and this inhibition of calling was enhanced following a training session in which the model stimulus was presented in association with grackle alarm calls. Acquired inhibition of calling was independent of the order of presentation of the model and an alarm chorus. These are the first results to indicate that social acquisition of predator avoidance is not dependent upon a particular temporal relationship between predators and social alarm cues. Evolution may have modified some properties of Pavlovian conditioning to accommodate social learning about potentially dangerous stimuli.

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It has long been recognized that learning should be 'adaptively specialized' for the function that it serves in nature (Rozin & Kalat 1971). What exactly qualifies a particular learning process as 'adaptively specialized' is, however, a subject of debate. For proponents of general process theory, most instances of learning arise from simple associations between novel stimuli (conditioned stimuli, CS), and biologically significant events (unconditioned stimuli, US). Adaptive specialization refers to the possibility that evolution has fine-tuned specific learning parameters to fit particular situations (Domjan 1980, 1983). Although some authors avoid referring to quantitative variation in learning parameters as adaptive specialization (Bolhuis & Macphail 2001; Macphail & Bolhuis 2001), the idea that learning processes, like other aspects of the phenotype, have been shaped by natural selection in response to the demands of particular environmental

situations is generally accepted by psychobiologists (Sherry & Schacter 1987; Shettleworth 1993, 1998, 2003; Kamil 1994; Gallistel 1999).

One potentially fruitful approach to understanding how functional demands may have shaped mechanisms of learning is to use modern associative-learning theory as a framework within which to compare learning phenomena. Such analyses may reveal that two instances of learning are mediated by the same mechanism, but are specialized in that the properties of learning have been modified to accommodate the need to learn about relationships between particular kinds of stimuli (Garcia & Koelling 1966). For example, although taste-aversion learning follows general laws of associative learning, taste-aversion learning is unique in that it is acquired in one trial even when gastrointestinal illness is induced hours after a food has been ingested (Domjan 1980).

Socially acquired predator avoidance has been found in fish, birds, and both eutherian and marsupial mammals. The pattern of acquisition is similar across groups. Although subjects initially show little or no response to a neutral stimulus, once the previously neutral stimulus has been presented together with an alarm signal, it

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evokes an avoidance response (reviewed in Griffin 2004).

Several authors have noted similarities between the process of predator avoidance learning and Pavlovian conditioning (Suboski 1990; Mineka & Cook 1993; Heyes 1994; Shettleworth 1998). Within this framework, the predatory cue is considered a conditioned stimulus to which observers acquire avoidance responses after the stimulus has been presented in contiguity with an alarmed demonstrator, the unconditioned stimulus. Discussion of learned predator avoidance as an instance of Pavlovian conditioning is supported by similarities in properties of social learning about predators and classical conditioning. For example, observer fear levels after training are positively correlated with those of the demonstrator during training (Mineka & Cook 1993). This positive relationship between the fear levels of demonstrators and observers is consistent with the observation that the strength of a classically conditioned response typically increases with the intensity of the reinforcer. Such findings have led to the broadly accepted view that socially acquired predator avoidance is mediated by Pavlovian conditioning, rather than any specialized learning process (Suboski 1990; Mineka & Cook 1993; Heyes 1994).

However, a recent review of the properties of social learning about predators (Griffin 2004) has revealed some differences between predator recognition learning and classical conditioning. In particular, prior exposure to a novel predator does not retard or inhibit the subsequent acquisition of an antipredator response to that predator (Curio et al. 1978; Mineka & Cook 1986). In contrast, in most instances, prior familiarity with a conditioned stimulus is known to delay acquisition of a classically conditioned response. The finding that effects of CS novelty on conditioning are different in learned predator avoidance than in other domains raises the possibility that the parallels between Pavlovian conditioning and socially acquired predator avoidance may be more limited than is generally thought.

There has been considerable debate about whether backward conditioning, a training procedure in which the delivery of the CS follows that of the US, rather than precedes it, supports acquisition of a response to a CS (Mackintosh 1974; Spetch et al. 1981; Hall 1984). According to traditional views, while forward conditioning, in which the CS is presented before the US, yields an acquired response to the CS, backward conditioning usually produces associations that are either transient or inhibitory. The traditional view, which is predicted by both associative (Rescorla & Wagner 1972; Mackintosh 1975; Pearce & Hall 1980; Wagner 1981; Pearce 1987), and several timing models of association learning (Sutton & Barto 1990; Grossberg 1991; Staddon & Higa 1999), is consistent with the assumption that the function of Pavlovian conditioning is to learn causal relationships between events because causes generally precede, rather than follow, their consequences. The traditional view is also supported by neurophysiological evidence of forward, but not of backward, neural conduction (Hammer 1993), a process presumed to underpin memory and learning.

The possibility that learning can only occur in forward relationships is not, however, consistent with a 'behaviour systems' framework (Timberlake 1994). This ecologically based approach assumes that behaviour patterns consist of an organized sequence of stimulus sensitivities and motor responses, and predicts that the nature of the conditioned response, rather than the occurrence of a conditioned response, will depend upon the temporal relationship between the US and CS (Silva et al. 1996, 1998). Occurrence of forward, but not of backward learning, is also contrary to predictions arising from the temporal encoding theory of animal learning (Matzel et al. 1988), a model that proposes that learning in backward preparations appears to be weaker than in forward conditioning, not because learning does not occur, but because learning is not properly assessed (Arcediano & Miller 2002). In this model, responses acquired in backward preparations are different from those acquired in forward preparations, so special procedures are needed to detect them (Arcediano & Miller 2002).

Most successful instances of backward conditioning have employed aversive USs (e.g. Wagner & Terry 1975; Mahoney & Ayres 1976; Tait & Saladin 1986; McNish et al. 1997; but see Hall 1984), a finding that has been attributed to the functional value of predator avoidance learning (Keith-Lucas & Guttman 1975; Spetch et al. 1981). For example, Keith-Lucas & Guttman (1975) found that a single foot shock followed by the presentation of a toy hedgehog (Erinaceus sp.) led to a robust acquired avoidance response of the hedgehog by rats. The authors attributed their success to use of a complex, biologically meaningful stimulus. However, this approach has not been pursued further.

Backward temporal relationships between predators and frightening events may be more common in a social context than when solitary individuals encounter predators. Especially in species whose members travel in groups, individuals should often detect the alarm behaviour of social companions before they detect a predator, and detection of the US before the CS should nevertheless be sufficient for predator recognition learning to occur. Furthermore, acquired avoidance responses should be identical whether a predator is detected before or after social alarm signals. It might, therefore, be predicted that social learning about predators should not be dependent upon the temporal relationship between detection of predator-related and social alarm cues (Galef 1988; Griffin 2004) and such learning should not (as Arcediano & Miller 2002 proposed) require special procedures to detect it. These predictions have not yet been tested. In fact, description of the temporal relationship of CS and US is often absent from experimental protocols of studies of predator avoidance learning (e.g. Curio et al. 1978; Vieth et al. 1980). The reader is left to assume that unfamiliar predator models and social stimuli are presented more or less simultaneously.

The carib grackle, Quiscalus lugubris, is a highly urbanized avian species common throughout the Caribbean Lesser Antilles. Grackles typically forage in small, mobile flocks and give high-amplitude, broadband pulsatile chuck vocalizations.

Several lines of evidence show that production of chuck calls is strongly associated with the presence of predators (Jaramillo & Burke 1999). In free-living grackles, high rates of chuck calls are typically evoked by mongooses, *Herpestes auropunctatus*, cats, *Felis catus*, vervet monkeys, *Chlorocebus aethiops*, and dogs, *Canis lupus familiaris*, and are sometimes accompanied by mobbing. In addition, experimental playback of chuck calls cause receivers to take flight and to begin calling (A. S. Griffin, unpublished data).

High rates of chuck vocalizations (average = 24.9 calls/min, range 5–50, $N = 10$) can be evoked experimentally in captive, individually caged grackles by a threatening stimulus, such as a human standing in front of the cage and visually tracking a target bird (A. S. Griffin, unpublished data). Given the strong association between chuck calling and the presence of potential predators, we refer to these calls hereafter as alarm calls.

The aims of the following two experiments were threefold. In experiment 1, we measured the response of carib grackles to a realistic model predator (a life-sized model of a fox terrier). These tests enabled us to determine whether trained antipredator responses obtained in experiment 2 resembled those evoked by a familiar predator, as has been shown in earlier studies of predator recognition learning (e.g. Curio 1988). In experiment 2, we determined whether paired presentations of a novel avian model (black-and-yellow pigeon) and grackle chuck calls enhanced the responses of grackles to the model bird. We also determined whether the likelihood of predator avoidance learning was influenced by the order in which the novel 'predator' and the alarm call sequences were presented.

GENERAL METHODS

Subjects and Husbandry

We caught carib grackles on the grounds of Bellairs Research Institute (St James, Barbados) using walk-in baited traps. We weighed each bird, individually identified it with a light-weight coloured metal leg band and placed it in an individual cage ($2.25 \times 2.25 \times 0.76$ m) in an outdoor aviary at the Bellairs Institute. At any one time, only three grackles were held in the aviary.

Cages were visually isolated from one another by black plastic sheets, and each was equipped with a branch perch (Fig. 1). Grackles had ad libitum access to water and a mixture of dog pellets and rice for the duration of the experiment. After testing, we released each bird back into the wild population from which we had captured it.

Acclimitization

We left the birds undisturbed for the first 48 h after capture to acclimatize to captivity, and all birds began to feed within a few hours of being placed in aviaries. On the third day after capture, the experimenter watched each individual once in the morning and again in the evening for 10 min from a blind 3 m from each bird's cage. Most birds fed while being watched, demonstrating that
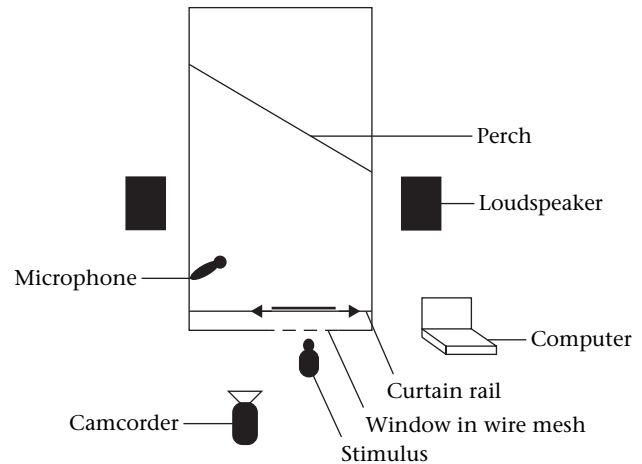

Figure 1. Plan view of an individual test cage. Each cage was equipped with a perch. Digital recordings of the acoustic stimuli were played back through two loudspeakers placed on either side of the cage and connected to a laptop computer. The visual stimuli were introduced into the cage through an opening in the mesh wire, which was hidden behind a curtain. A microphone connected to a digital camcorder was used to record the alarm call responses of the subject grackle to each visual stimulus (see text for more details).

presence of the experimenter did not seriously disturb them. Our subjects may have adjusted particularly rapidly to captivity because the grackle population around Bellairs Institute lives in proximity to humans, and the birds are accustomed to being fed. Experiments began on the fourth day after capture.

All husbandry and experimental procedures used here were described in Animal Utilization Proposal No. 4660 and approved by the McGill University Animal Research Ethics Board.

EXPERIMENT 1

Earlier studies of predator avoidance learning have shown that trained antipredator responses are similar to those evoked by a natural predator (e.g. Curio 1988). The aim of experiment 1 was to quantify the responses of grackles to a model of a familiar predator, a dog, so that in experiment 2 we could assess qualitatively the resemblance between trained and spontaneous (i.e. nontrained, antipredator responses.

In the absence of any apparent disturbance, particularly in the early morning and late afternoon when grackles are most active, individually caged captive grackles give chuck vocalizations at low rates (average = 8.9 calls/min, range 0–28, $N = 10$). Free-living solitary grackles also call at low rates, typically after landing at a food source and before beginning to forage.

Interestingly, some events suppress baseline calling in captivity. For example, grackles tend to go quiet if a human enters the aviary without looking at the birds. Similarly, pilot trials suggested that presentations of model predators inside the aviary suppressed baseline alarm calling. This inhibition of chuck calling contrasted with observations that captive birds increase chuck calling rates in response

to live predators, such as cats and dogs, located more than 10 m from the aviary (A. S. Griffin, personal observation), suggesting that, like many other animals, grackles may vary their alarm-calling rates as a function of the distance to a predator (Curio & Regelmann 1985; Pereira & Macedonia 1991; Blumstein 1995). To better understand whether predator presentations enhance or inhibit chuck calling in captive carib grackles, we presented each bird with a model dog both inside the cage and 3 m away from the aviary.

Methods

Subjects

Ten adult carib grackles (one female, nine males) served as subjects in experiment 1.

Test procedure

We conducted one stimulus presentation per day in the early morning. Before each test, we hung a curtain over the front of each cage adjacent to that containing the focal grackle. In this way, we ensured that only the focal subject saw the test stimulus.

The predator stimulus was a realistic, life-sized (0.4 × 0.3 × 0.15 m), brown and white, furry, fox-terrier toy. We presented each grackle, in random order, for 60 s, with the predator model both inside the cage and 3 m away from its front wall.

At one end of the cage, a curtain screened an opening (0.2 × 0.2 m) in the wire mesh (Fig. 1). To present the dog inside the cage, the experimenter sat behind a hide placed 3 m away from the enclosure, and operated a system of strings to open the curtain and a 3-m-long stick to push the model dog out of a black box in which it was hidden and into the cage. At the end of the presentation, the experimenter pulled on the stick to withdraw the dog from the cage and into the box from which it had emerged, and operated the system of strings to close the curtain. During tests in which the model predator was presented at a distance of 3 m from the cage, the experimenter reached through a slit in the plastic of the hide and placed the stimulus on a stool in front of the hide. A wall alongside the aviary made it impossible to test the effects of presenting the predator stimulus at distances greater than 3 m.

Alarm vocalizations

Adjacent cages were separated by approximately 1 m, making it difficult to discriminate between a focal subject's chuck calls and those of its two neighbours. Consequently, we placed a Sony dynamic F-V620 microphone at one end of the cage and connected it to a Sony Digital 8 camcorder (Fig. 1). The focal bird's chuck calls were much louder on the recordings than those of the two other grackles in the aviary, and could be counted easily. We videorecorded the grackles for 1 min immediately before predator presentation (baseline), 1 min during predator presentation, and 3 min after the predator had disappeared. To quantify responses to the predator, we measured changes in alarm-calling rates from baseline. For each bird, we counted the number of chuck calls produced during the 1-min prestimulus baseline and each of the 1-min intervals after appearance of the predator model. We then calculated the difference in number of chuck calls between baseline and each time interval after the appearance of the model predator.

We examined effects of presentation distance on chuck-calling rates by using a two-way, repeated measures ANOVA with factors for distance (close, far) and time (four 1-min time intervals). Significance levels were set at 0.05 and all tests were two tailed. All analyses were carried out on untransformed data using Statview 5.2 (SAS Institute 1998) and Superanova 1.1 (Abacus Concepts 1991).

Results and Discussion

Grackles decreased their chuck calling significantly more when the model predator was presented inside their cage than when the predator model was presented 3 m away from the enclosure (Fig. 2). Formal comparisons between responses to the model dog presented inside the aviary (close) and responses to the same stimulus presented 3 m away (far), revealed a significant distance by time interaction ($F_{3,27} = 8.620$, $P < 0.001$). Main effects of distance and time were not significant (distance: $F_{1,9} = 2.900$, $P = 0.123$; time: $F_{3,27} = 2.691$, $P = 0.066$).

The finding that grackles decrease alarm calling in response to a predator presented at close range is perhaps surprising. However, it is consistent with other studies that have examined the likelihood of alarm signalling as a function of distance to a predator. For example, Thomson gazelles, *Gazella thomsonii*, are less likely to stot when a predator is close (Caro 1986). Ringtailed lemurs, *Lemur catta*, chased by a dog flee silently until they reach the safety of a tree branch, and only then, do they begin calling (Pereira & Macedonia 1991). Golden marmots,
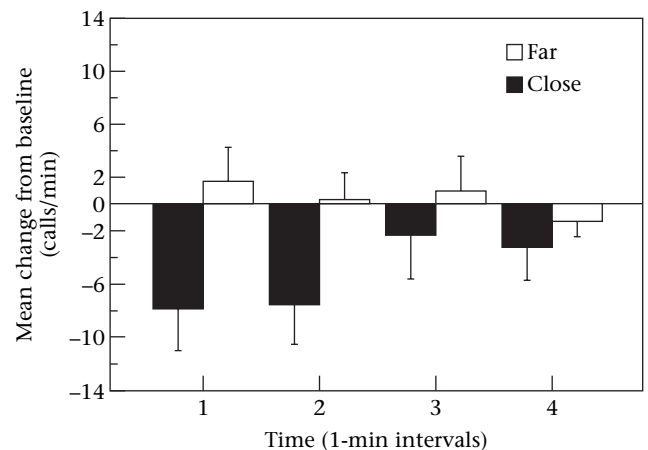

Figure 2. Changes in alarm-calling rates from prestimulus baseline in response to a model dog presented inside the cage and 3 m away from the aviary. The mean + SE change from baseline ($N = 10$) is plotted for four successive 60-s time intervals from stimulus onset. This period includes the stimulus presentation (1 min) and three 1-min postpresentation intervals.

Marmota caudata, give alarm calls with decreasing numbers of notes per call as a human approaches, then suppress alarm calling altogether and run to their burrow when the human comes close (Blumstein 1995).

In birds, studies of alarm calling usually show that calling rates increase with decreasing distance to a predator (e.g. Curio & Regelmann 1985). However, because in most studies callers are free to fly away, they do not remain close to the predator for long. Suppression of alarm calling might be seen if flight were restricted. Indeed, Curio (1993) reported that birds held in an aviary respond to predator models with antipredator behaviour, while those held in cages suppress antipredator responses. It is also well known that birds under immediate risk (e.g. a raptor in flight nearby) vocalize relatively little (Gyger et al. 1987). Together these studies show that animals may refrain from alarm signalling when they are at high risk of predation. Our grackles may have suppressed chuck calling in the presence of the model dog because they perceived the model as an imminent predatory threat.

EXPERIMENT 2

Experiment 1 demonstrated that presentation of a model of a familiar predator caused captive grackles to decrease the rate at which they called. In experiment 2, we determined whether presentations of a previously unfamiliar avian model (a black-and-yellow pigeon) together with chuck calls led to acquisition of an antipredator response qualitatively similar to that observed in the presence of a familiar predator. We also tested whether order of presentation of the novel stimulus and playback of alarm calls affected the likelihood of learning to suppress alarm calling in the presence of an unfamiliar model.

It is relevant to note that there is disagreement in the literature as to what constitutes a backward pairing. According to Kimble's (1961) classification scheme, all procedures in which the CS onset follows US onset are defined as backward. Other authors restrict the definition of backward pairings to those in which US offset occurs before CS onset (e.g. Arcediano & Miller 2002). We elected to follow Kimble's (1961) definition and to use a backward design in which the social alarm chorus began before the appearance of the novel 'predator' and overlapped with it. We chose this design because we considered it most similar to a natural encounter with a predator. It is unlikely that, in natural circumstances, a group of alarm-calling individuals would fall silent when one of its members first sees a potential predator.

Birds in a forward conditioning group were presented with the model pigeon 20 s before onset of a 3-min alarm chorus playback. Thus, for birds assigned to the forward conditioning group, the appearance of the pigeon predicted onset of the alarm sequence. According to associative-learning theory (Rescorla 1988), this CS–US temporal arrangement should produce a robust acquired antipredator response to the novel stimulus.

Individuals in a backward conditioning group were shown the model pigeon 20 s after onset of alarm chorus playback. In this group, appearance of the pigeon was associated with the alarm sequence, but did not predict it. If the properties of social learning about predators are similar to those of Pavlovian conditioning, grackles receiving the backward conditioning treatment should not acquire an antipredator response to the pigeon. In contrast, if social learning about predators is not dependent upon predictive relationships between predators and social alarm cues, as functional considerations predict, then grackles assigned to the backward conditioning group should acquire the same antipredator response to the pigeon as those assigned to the forward conditioning group. Finally, to control for any changes in behaviour that occurred as a consequence of nonassociative learning (Rescorla 1967), grackles in an unpaired control group experienced the predator and the alarm playbacks, but the two events were widely separated in time.

To isolate effects of training, we measured focal subjects' responses to the model pigeon both before (pretraining test) and after (post-training test) training. Comparisons between forward and unpaired control groups, and backward and unpaired control groups enabled us to isolate effects specifically attributable to associative learning and to separate such effects from those that might be a consequence of nonassociative factors, such as confinement and repeated exposure to conspecific alarm calls (Shettleworth 1998).

Methods

Subjects

Twenty-four adult carib grackles (six females, 18 males) were used in experiment 2. None of these subjects had taken part in experiment 1.

Visual and acoustic stimuli

Because familiarity with a stimulus can interfere with subsequent acquisition of responses to that stimulus, we selected a CS that was unfamiliar and had no resemblance to natural predators of grackles (Curio 1988), a model bird, of the shape and size of a common urban pigeon with a 0.15-m-long tail, and painted black with a yellow breast.

To create the alarm call playback sequences, we made several recordings of chuck vocalizations of two captive carib grackles. Vocalizations were recorded and digitized using a Sony dynamic F-V620 microphone connected to a G3 iBook computer (Amadeus sound software, sample rate 44.1 kHz, 16-bit amplitude encoding). We then edited the recordings to make two distinct 180-s alarm call choruses. Within each chorus, we simulated the effect of grackles calling from multiple locations by playing back one series of calls through the right channel and another through the left channel of the stereo stimulus. To avoid startling the birds, each chorus began with a 4-s fade-in (0–87 dB) and ended with a 4-s fade-out (87–0 dB). It was played back at an amplitude of 87 dB (A weighting; peak; ± 1 dB measured 1 m in front of the speaker) roughly equivalent to the birds' own output volume measured at a distance of 2 m. We played back the choruses using an iBook G3 computer through two Altec Lansing 220

speakers, which we hid behind the black plastic on each side of the cage.

Test procedure

Each bird first received a pretraining trial during which we determined its initial response to the model pigeon. We then conducted one training trial in which each grackle received paired (forward and backward groups) or explicitly unpaired (control group) exposure to both the model pigeon and a randomly selected alarm call chorus. We conducted pretraining tests early in the morning and training trials 1.5–3 h after the pretraining tests. Post-training tests were conducted the following morning.

During pre- and post-training trials, and during training trials, we introduced the model pigeon into the subject's cage using a procedure identical to that used to present the model dog in experiment 1. During pre- and post-training trials, we presented the model pigeon for 60 s. For training trials, we presented the model pigeon either 20 s before (forward conditioning group) or 20 s after (backward conditioning group) the onset of the 180-s alarm chorus playback. We withdrew the pigeon from the cage either 40 s before (forward conditioning group), or simultaneously with the end of the alarm chorus (backward conditioning group). Total presentation time was thus 160 s for both the forward and the backward conditioning group. We presented grackles in the explicitly unpaired control treatment with the pigeon for 160 s and with the 180-s alarm chorus, but these two events were separated by a minimum of 40 min and a maximum of 2 h, and their order of presentation was balanced across birds.

During all tests, grackles in the adjacent cages were visually, but not acoustically, isolated from the focal bird and the experimental stimuli.

Alarm vocalizations

We videorecorded all trials, but scored and analysed only pre- and post-training tests. During pre- and post-training trials, we videorecorded each bird for 1 min immediately before presentation of the model pigeon (baseline), 1 min during the model presentation, and 3 min after the stimulus had disappeared.

To quantify responses to the model pigeon, we measured changes in levels of calling from the prestimulus baseline. To do so, for each grackle, we counted the number of chuck calls produced during the 1-min prestimulus baseline, the 1-min stimulus presentation, and the 3-min period after the predator model had disappeared. For the 3-min postpresentation period, we calculated the mean number of calls per min. We then calculated the difference in number of chuck calls between baseline and each time period (presentation, postpresentation).

First, to ensure that any differences between the post-training responses of subjects assigned to the three treatments were not due to any differences in their initial responses to the pigeon stimulus, we compared the mean change from the baseline rate of calling of the three groups during pretraining tests using a two-way, repeated measures ANOVA with factors for group (backward, forward, unpaired) and time (presentation, postpresentation).

Second, to identify the effects of training, we compared the mean change from baseline rate of calling in the pretraining tests with that of post-training tests using a three-way, repeated measures ANOVA with factors for group (backward, forward, unpaired), test (pretraining test, post-training test) and time (presentation, postpresentation).

Third, to compare the mean acquired responses of the backward and forward groups with that of the unpaired control, we collapsed the time variable (presentation, postpresentation) and calculated the difference between the pretraining response and the post-training response for each bird. We then calculated the mean difference for each group. We compared the values of the experimental groups (backward, forward) with that of the unpaired control using independent contrasts (Abacus Concepts 1991).

We used an alpha level of 0.05 throughout, except for contrasts. For contrasts, we used a corrected alpha level of 0.025 to account for the test of two successive comparisons (Bonferroni 1937). All tests were two tailed. All analyses were carried out on untransformed data using Statview 5.2 (SAS Institute 1998) and Superanova 1.1 (Abacus Concepts 1991).

Results and Discussion

Before training, the grackles' responses to the model pigeon did not differ significantly between groups (main effect group: $F_{2,21} = 1.574$, $P = 0.231$; main effect time: $F_{1,21} = 2.956$, $P = 0.100$; group∗time interaction: $F_{2,21} = 1.892$, $P = 0.176$; Fig. 3). After training, grackles that had seen the pigeon appear before the onset of the alarm chorus playback (forward group), and those that had seen the model appear after the onset of the chorus (backward group), decreased alarm calling in response to the model
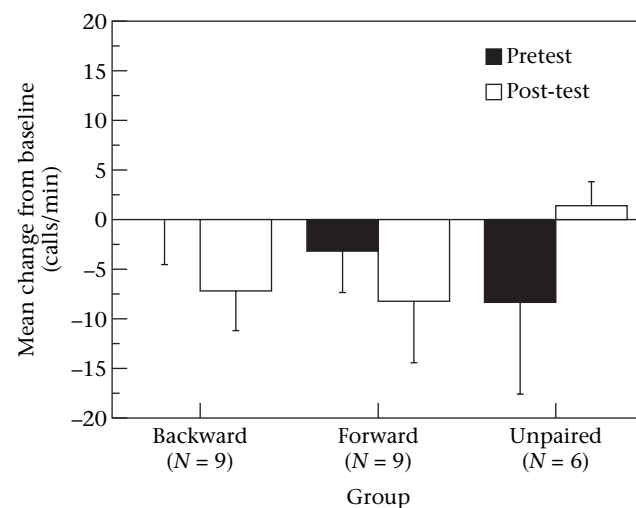

Figure 3. Alarm-calling responses evoked by a model pigeon both before (pretraining test) and after (post-training test) training. The mean + SE change from prestimulus baseline was averaged across the 1-min stimulus presentation period and three 1-min postpresentation intervals for each group and each test.

bird more after training than before training (Fig. 3). In contrast, the control group that had experienced no relationship between the pigeon and the alarm chorus during training did not decrease alarm calling in response to the model after training relative to before training (Fig. 3).

The differential effect of paired versus unpaired training sessions was reflected by a significant interaction of group∗test ($F_{2,21}$ = 4.360, P = 0.026; Fig. 3). There were no significant main effects of group ($F_{2,21}$ = 0.283, P = 0.756), or test ($F_{1,21}$ = 0.113, P = 0.740). In contrast, there was a main effect of time ($F_{2,21}$ = 5.051, P = 0.036), reflecting the fact that grackles in all groups and all tests suppressed calling more while the model pigeon was in the cage than after it had disappeared. None of the three-way interactions, or interactions involving time, were significant.

Planned pairwise comparisons showed that the grackles for which the appearance of the model bird signalled the onset of the alarm chorus decreased chuck calling after training significantly more than did grackles assigned to the unpaired control group (forward versus unpaired groups: $F_{1,13}$ = 5.908, P = 0.024), demonstrating that they had learnt that the appearance of the pigeon predicted the alarm calls. Grackles in the backward conditioning group, for which the model bird appeared only after the onset of the chorus, also decreased alarm calling significantly relative to the unpaired control (backward versus unpaired groups: $F_{1,13}$ = 7.838, P = 0.011), indicating that subjects in this group had also learnt to associate the pigeon with the alarm calls, despite backward presentation of the two stimuli. The pattern of results demonstrates that changes in behaviour in both the forward and backward treatments were attributable to associative learning and not to nonassociative factors, such as confinement in captivity or exposure to social alarm calls. Most importantly, these results demonstrate that learning was independent of the temporal relationship between presentation of the novel 'predator' and social alarm cues.

It may seem surprising that social avoidance training produced decreases in alarm-calling rates. Results from experiment 1, however, demonstrated that, under our experimental conditions, grackles decreased alarm calling in response to a familiar predator presented at close range. The fact that grackles decreased alarm-calling rates further after training than before training is, therefore, consistent with results of earlier studies of predator avoidance learning in birds that have found that responses acquired as a consequence of training are qualitatively similar to those evoked by a familiar predator (Curio et al. 1978; Maloney & McLean 1995).

Unfortunately, most studies of social learning of response to predators encode acquired responses as an index of disturbance, which incorporates weighted averages of both vocal and nonvocal antipredator responses (Curio et al. 1978; Vieth et al. 1980; Maloney & McLean 1995; Hölzer et al. 1996; van Heezik et al. 1999). Consequently, it is not possible to evaluate exactly how alarm calling per se changed as a consequence of training and direct comparisons with the present results are difficult.

In conclusion, our results demonstrate that both forward and backward relationships between a novel stimulus and social alarm cues produced robust positive associations between the two events. In addition, the similarity between the responses acquired as a consequence of training and those evoked by a familiar predator lead us to conclude that our training regime mimicked an instance of predator avoidance learning and that the acquired response reflected an increase in predator avoidance.

GENERAL DISCUSSION

Presentations of an unfamiliar avian model (black-and-yellow pigeon) caused carib grackles to decrease the rate at which they produced alarm calls, as did presentations of a model of a familiar predator. The effect of the model pigeon on calling rate was enhanced after one training session in which the avian stimulus was presented together with a playback of grackle alarm calls, suggesting that, as a consequence of training, the grackles became more wary in the presence of the avian stimulus. Whether the novel 'predator' was presented before or after the onset of an alarm call chorus did not affect the likelihood of associative learning.

These results extend the range of avian species that are known to show socially acquired predator avoidance (reviewed by Griffin 2004), and provide the first evidence of such learning in a New World blackbird (Icteridae). More important, these findings are the first to demonstrate that the social acquisition of predator avoidance is not dependent upon a forward relationship between a novel predator (CS) and a social alarm cue (US), as would be expected given the relatively high probability of the onset of alarm calling (US) preceding observation of a predator (CS) by a naïve individual in natural situations.

According to some definitions, but not others, presentations in which alarm signals both precede and overlap presentation of a novel predator constitute backward presentations. We chose such a design because it was most similar to our field observations of encounters between predators and alarm-calling groups of potential prey, and is consistent with classification schemes that define as backward procedures all those in which the US onset occurs before CS onset. Although we cannot exclude the possibility that stimulus synchrony played a role in the acquisition of an avoidance response to the pigeon, our results demonstrate that within the limits tested, social learning about predators is not affected by temporal relationships between presentation of social alarm stimuli and a predator. This finding contrasts with conditioning involving other kinds of stimuli, such as tones and electric shocks, in which the likelihood of learning is sensitive to CS–US timing (Smith et al. 1969). Our work is an important first step towards understanding the role of timing in social learning about predators, a learning context in which functional considerations predict that temporal flexibility should be favoured. Although an experimental design with no overlap between social alarm stimuli and predator seems unlikely to resemble a natural situation, it

will be interesting to determine whether predator recognition learning occurs under those conditions.

According to contemporary views, associative learning allows an animal to learn about relationships between causes and effects; associations between two events are learnt not because events occur in close temporal contiguity, but because occurrence of one predicts occurrence of the other (Rescorla 1988). A direct consequence of this view is that animals should learn to respond to stimuli that precede, rather than follow, a biologically significant event. Backwards learning is, therefore, particularly challenging to explain.

A mix of successful and unsuccessful attempts to produce positive backward associative learning has fuelled an ongoing debate surrounding the reality of the phenomenon (Spetch et al. 1981; Hall 1984; Miller & Barnet 1993; Silva et al. 1998; Arcediano & Miller 2002). Backward conditioning is typically explored using lights and tones as signals for delivery of food or electric shock. Divergent results in studies of backward conditioning are attributed to lack of control groups (Hall 1984), to procedural differences (Wagner & Terry 1975; Albert & Ayres 1997), to choice of measures of acquired responses (Mahoney & Ayres 1976; Tait & Saladin 1986; McNish et al. 1997; Arcediano & Miller 2002), or to formation of underlying forward and/or simultaneous associations (Romaniuk & Williams 2000; Williams & Hulburt 2000).

One attempt to explain divergent results in studies of backward conditioning has been to suggest that learning in backward preparations can only be detected using particular experimental procedures (Arcediano & Miller 2002), because responses expressed in anticipation of an event are different from those displayed after the event has occurred. Consequently, tests of learning that measure anticipatory responses will reveal little or no acquired response following backward conditioning (Matzel et al. 1988; Arcediano & Miller 2002).

Another attempt to reconcile the acquisition of backward relationships with theory has been to suggest that the ability to learn backward associations evolved as an adaptation to learning about predators (Keith-Lucas & Guttman 1975; Spetch et al. 1981). In such a theory, backwards learning may occur less reliably in experimental paradigms such as conditioned eye blinking and appetitive conditioning, in which backward relationships have little functional significance (but see Lehrer 1971; Silva et al. 1996, 1998).

Although fear conditioning might tap into predator avoidance learning, backwards relationships between fear and predators are unlikely to be common in direct interactions between prey and predators, because prey would need to survive a surprise attack by a predator to experience fear. In contrast, backwards relationships between social alarm cues and predators might be the rule rather than the exception. Consequently, we propose that backward learning might be a property specific to social learning about predators.

In summary, there is little doubt that socially acquired predator avoidance is mediated by associative learning in which novel stimuli are linked to social alarm cues (Heyes 1994). The finding that social learning about predators is not strictly dependent upon the order in which social alarm cues and predator are detected can be explained within the general process approach to the study of learning, by using a subset of existing learning theories (e.g. Matzel et al. 1988; Gallistel & Gibbon 2001). The present finding of irrelevance of the order of onset of CS and US in socially learned predator recognition can also be explained within the ecological approach to the study of animal learning, by suggesting that evolution may have modified some of the properties of Pavlovian conditioning to fit functional demands, as has been found for other instances of learning (Domjan 1980). Which of these explanations is more appropriate will become clear only after consensus is reached as to how associations arise and further work on the importance of temporal relationships between CS and US in social learning about predators has been completed.

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