The reaction of wild-caught northern brown bandicoots (*Isoodon macrourus*) to predators

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Running head Bandicoot behaviours towards predators

Short summary

Some Australian mammals may not recognise introduced predators, and are naïve to the threat they pose. We assessed how wild-caught bandicoots reacted to a variety of predators, and our results show that bandicoots may recognise a live dog or cat as a threat. Bandicoots may need to be trained to recognise and respond appropriately to some predator cues to maximise their chance of survival in the wild.

Abstract

Australia's mammals are facing devastating effects from introduced predators such as cats and foxes. It is hypothesised that this may be because Australian mammals do not recognise some predators, and are naïve to the threat they pose. We tested how wildcaught northern brown bandicoots (*Isoodon macrourus*), reacted to a variety of predators and their cues in captivity. These cues were live predators, taxidermied predators, and predator faeces. Bandicoots spent less time feeding and took longer to begin feeding in the presence of a live dog. However, they did not display a significant difference in behaviours in the presence of any other predator cue when compared to the baseline. This suggests that bandicoots may perceive live dogs as a threat , and the use of taxidermied predators and predator faeces may not elicit avoidance behaviours in bandicoots. Bandicoots may need to be trained to recognise and respond appropriately to some predator cues to maximise their chance of survival.

Additional keywords – bandicoot; predator-prey; naïvety; prey naïvety; marsupial; predator-prey interactions

Introduction

Australia has the highest mammal extinction rate in the world, with at least 27 mammal extinctions recorded since European settlement (Woinarski *et al.* 2015; Kearney *et al.* 2019). The introduction of cats (*Felis catus*) and foxes (*Vulpes vulpes*), in particular, has led to the decimation of native wildlife populations. During European settlement, the Australian landscape went through rapid restructuring, with unsustainable farming practices leading to overgrazing, land clearing, and altered fire regimes (Johnson 2006; Woinarski *et al.* 2015). This clearing of the landscape resulted in reduced concealment opportunities for native animals, and prime hunting conditions for introduced predators, amplifying the effects of these predators (Johnson 2006).

Some of Australia's mammals are also said to be naïve to introduced predators (Cox and Lima 2006; Banks and Dickman 2007; Banks *et al.* 2018). Thus, some Australian mammals display relaxed predator avoidance behaviours towards predators, and therefore do not recognise the threat these introduced predators pose, or exhibit inappropriate antipredator behaviours (Cox and Lima 2006; Banks and Dickman 2007; Banks *et al.* 2018). Consequently, this field of research is growing, with naivety experiments conducted on a variety of species (Banks *et al.* 2018; Edwards *et al.* 2021).

Whilst odours (from faeces, integument, or urine) are the most commonly used cues in naivety assessments (Edwards *et al.* 2021), predator odours such as from faeces may not be indicative of a real threat of being killed, as the likelihood of an encounter with a predator at a scat is low (Banks *et al.* 2003). Thus, investigation is needed into the suitability of predator odour (e.g. from faeces, urine, integument) presented to native animals during testing, and the repeatability of such tests. Similarly, investigation into the suitability of taxidermied animals or models of animals (e.g. McLean *et al.* 1994), and live predators as a cue to test recognition is warranted, including the practicalities of using such cues.

Knowledge of how animals respond to predators and their cues can aid in determining if they are a good source population for translocations (Bannister *et al.* 2018), how varying predator densities may impact them (Moseby *et al.* 2019), the impact of newly introduced predators into an area (such as the introduction of foxes to Tasmania) (Jones *et al.* 2004), and whether predator avoidance training may be useful during translocations or reintroductions (Moseby *et al.* 2016).

One group of animals that have faced particularly devastating effects from introduced predators are the bandicoots (family: Peramelidae). Since European settlement of Australia, three species have become extinct (Burbidge and Woinarski 2016a, 2016e, 2016g), four species are classified as Vulnerable (Burbidge and Woinarski 2016b, 2016d, 2016f; Woinarski and Burbidge 2016), and four species are classified of Least Concern (Burbidge and Woinarski 2016c; Leary *et al.* 2016; Lunney *et al.* 2016a; Lunney *et al.* 2016b). Given their heightened vulnerability to introduced predators, knowledge of how bandicoot species respond to predators and different predator cues may aid in conservation efforts such as translocations and reintroductions.

Three bandicoot species have previously been examined for recognition of both native and introduced predators, using either trap success in the presence of predator faeces (Russell and Banks 2005; Mella *et al.* 2010), or presence of diggings in backyards with and without pets to assess recognition (Carthey and Banks 2012; Frank *et al.* 2016). However, no studies have investigated how bandicoots behave with a variety of predator cues, including direct observation of behaviours in the presence of live predators.

We investigated the behavioural responses of wild-caught northern brown bandicoots (*Isoodon macrourus*) housed in captivity to three predator cues: faeces, taxidermied animals, and live animals. Our aim was to determine if bandicoots displayed different behaviours when in the presence of a predator cue, including their: 1) activity time budget (e.g. duration of time spent feeding); and 2) their latency to feed. We hypothesised that if bandicoots recognised the predators as a threat, they would increase vigilant behaviours and increase their latency to feed in the presence of a predator or their cue. Differences between these three cue types are discussed, along with implications for further studies in this field of research.

Methods

Study Species

We captured 15 wild-caught northern brown bandicoots (hereafter bandicoots), using baited cage traps in south-east Queensland (Table 1). Bandicoots are an omnivorous, ground-dwelling species, that prefer low groundcover, and weigh between 1-3 kg (Gordon 2008). This species of bandicoot is considered common in Queensland but threatened by introduced predators (Lunney *et al.* 2016b). They were chosen as a model species due to their similar niche occupation to threatened bandicoot species, and because they fall within the critical weight range of mammals threatened by introduced predators in Australia.

Housing

Bandicoots were housed at the Hidden Vale Wildlife Centre, located near Grandchester in south-east Queensland, Australia. Enclosures were 7 m wide x 6 m high x 12 m long on a natural soil substrate with a range of introduced and native plants including grass tussocks, and logs and piles of rocks. The setup of each enclosure was consistent to reduce variability. The bandicoots were fed a maintenance diet consisting of fruit, vegetables, live food (such as mealworms), oats and dog food (Jackson 2007). Bandicoots were initially trained to use microchip-automated nest boxes (Edwards *et al.* 2020), as a method of accessing safe refuge. Once the bandicoots had been trained to use the nest boxes, each bandicoot was randomly assigned to either the odour, taxidermied animal, or live animal treatment group, resulting in five per group. Baseline behavioural activity was recorded for three days for each bandicoot, to create mean baselines of behaviours for each treatment group. Recognition tests using the different predator cues then began after this baseline period. All bandicoots were eventually returned to the wild after the experiments.

Odour Recognition Tests

Bandicoots in the odour treatment group were presented with two feeding trays filled with sand (to a depth of approximately 2 cm) with their standard feed bowls placed on top. Their food ration was weighed and divided equally between the two trays. During the baseline period, both trays were filled with clean sand. During testing, on one tray, either: fox (*Vulpes Vulpes*), cat (*Felis catus*), macropod (eastern grey kangaroo, *Macropus giganteus*) (native non-threatening), or python (carpet python, *Morelia spilota*) (native predator) faeces (approximately 9-10 g) were placed on top of the sand (test tray). The other tray had no faeces (control tray). Each odour type had individual sand and trays to prevent odour cross-

contamination. Fox and wallaby faeces were provided freshly collected from the field. The cat and python faeces were provided by private owners. All faeces were frozen and thawed to room temperature before use.

Two testing sessions occurred per night, once when the bandicoots first emerged, and again at a random time at least 30 mins after the first session. In each session, the trays were placed in the enclosure, approximately 2 m apart. After a period of 10 mins from when the bandicoot first emerged for feeding, the trays and food were removed. Remaining food was placed back in the bandicoot's enclosure at the end of the testing. Each bandicoot was exposed to each odour, with different odours used on separate nights in a randomised order. This approach in presentation occurred for all odour, taxidermied animal, and live animal tests.

Taxidermied Animal Recognition Tests

Bandicoots in this experimental group were exposed to a taxidermied animal: either a cat, fox, or macropod (brush-tailed rock wallaby, *Petrogale penicillata*) (native non-threatening). Two testing sessions occurred per night, as described above. In each session, the taxidermied animal was placed in the top corner of the enclosure (just out of view of the camera), approximately 1.5 m from the bandicoot's singular food bowl.

Live-animal Recognition Tests

Bandicoots in the live-animal treatment group were exposed to a live animal: either a cat, a dog (*Canis lupus familiaris*), or a guinea pig (*Cavia porcellus*) (non-threatening). A dog and a guinea pig were used in replacement of a fox and wallaby respectively in this treatment group given the logistics of acquiring and using live wildlife. Three dogs were used: one in replicates one, three and four; one in replicate two; and one in replicate five. Two of the dogs were Border Collies of similar weight (~15-18 kg), build and colour. The third dog was a similar build and weight (18 kg), but a different breed (a Husky-mix) and a different colour to the Border Collies. The cat was black and weighed approximately 5 kg. Two different guinea pigs were used; both were short-haired and weighed approximately 1 kg.

Two testing sessions occurred per night, as described above. In each session, the live animal was taken into the top corner of the enclosure (just out of view of the camera) with a handler (the same handler for all animals in all replicates), approximately 1.5 m from the bandicoot's

singular food bowl. During the baseline period, the handler stood in the same location in the enclosure, to ensure that any differences observed between the treatments and the baseline were not a result of the human handler.

Behavioural Activity and Analyses

Infrared remote surveillance cameras (Kobi, Taiwan) were set to continuously record during the 10-min tests to collect focal animal all-occurrence behavioural activity in each enclosure. Video footage was later viewed, and behaviours recorded blind to the predator type where possible to reduce potential for observer bias. Footage was coded between 0.25 and 1 x speed to ensure accuracy. Behavioural activity of the bandicoots was categorised using a partial ethogram (Table 2). Variables investigated included: 1) latency to feeding (i.e. how long until feeding occurred during the 10-min test), and 2) duration of behaviours.

Given the small sample sizes, we used paired t-tests to compare the mean duration of behaviours for each bandicoot to their baseline mean, for each treatment group to determine if they differed significantly. A Bonferroni adjustment was made to minimise the chance of Type 1 errors (Rice 1989) and α was reduced to $P \leq 0.016$.

Permits

We conducted this research under Scientific Purposes Permits from the Department of Environment and Science (WA0001464; WIEP12989713; and WA0007908). Animal Ethics approval was obtained from The University of Queensland's Native/Exotic Wildlife and Marine Animals Ethics Committee (SAFS/126/17 and ANRFA/SAFS/336/17).

Results

For the odour treatment group (Fig. 1) and the taxidermied animal treatment group (Fig. 2), no behaviours statistically differed from the treatment baseline means. Bandicoots spent significantly less time feeding (P=0.008) and took longer to begin feeding (P=0.014) when in the presence of a live dog when compared to the baseline means. No other behaviours statistically differed from the baseline means for the live animal treatment group (Fig. 3), however there tended to be a similar pattern in behaviours by bandicoots when exposed to the live cat, though these results were not statistically significant.

Discussion

Bandicoot behavioural responses

Bandicoots showed a greater response to live predators than the predator faeces or taxidermied predators. During the odour experiment using predator faeces, bandicoots tended to have a shorter latency to feed (i.e. started to feed quicker) when in the presence of cat or fox faeces, and did not increase vigilance behaviours, although these results were not significant. Similar results have been found with cat-naïve bettongs, which did not increase vigilant behaviours in the presence of predator scents (Saxon-Mills et al. 2018). This indicates there may be a lack of wariness or recognition of the predator faeces as potentially threatening in this context. The bandicoots may not have reacted to the predator faeces because in the wild, as encounters with faeces may not be indicative of an encounter with an actual predator, and therefore may not signal a real threat (Banks et al. 2003). Similarly, detection of predator faeces may not elicit a 'typical' predator response such as fleeing (Tay et al. 2021), and was therefore unable to be detected in this study. This may explain why there were no significant changes in behaviours displayed by bandicoots when in the presence of python faeces, which as a coevolved native predator, could be expected to elicit a response. The use of integument odour (such as fur or skin) may be a more appropriate odour to test the naivety of species in future experiments, as it has been shown to provoke a stronger response than faecal odours (Blanchard et al. 2003; Apfelbach et al. 2005; Garvey *et al.* 2016).

When in the presence of the taxidermied animals, the lack of significant change in display of any behaviours indicates that the bandicoots did not recognise the taxidermied cat or fox as a threat. When in the presence of a live cat or a live dog, bandicoots generally spent less time feeding, took longer to start feeding . This suite of behavioural changes may indicate wariness and possible recognition of cats and dogs as a threat, with the response to dogs generally greater than to cats. These results support the notion that bandicoots, like other Australian mammals, have evolved antipredator behaviours towards dogs (Carthey and Banks 2012; Banks *et al.* 2018; Edwards *et al.* 2021). This may be because Australian fauna has a far longer history of co-occurrence with dogs (introduced between 3,500 and 5,000 years ago) than cats or foxes, which were only introduced in the last 220 years (Savolainen *et al.* 2004; Johnson 2006; Woinarski *et al.* 2015; Balme *et al.* 2018).

While typically an increase in vigilance may suggest recognition of a predator (Saxon-Mills *et al.* 2018), the vigilance behaviours recorded were generally short in duration, and may

indicate general wariness or investigation rather than recognition of a known threat. Indeed, bandicoots seemed to respond to a threat by spending more time out of view and away from the predator (as with the live dog), which is likely a more suitable reaction when threatened by a predator.

It is possible that a physiological stress response to predators was occurring, despite an absence of significant behavioural changes in most tests. Faecal cortisol is a non-invasive technique that could be used to measure a stress response to the predators (Dowle *et al.* 2013; Narayan *et al.* 2013). However, this was considered beyond the scope of this study, and would have entailed logistical difficulties such as finding the bandicoots' faeces in the large naturalistic enclosures.

Limitations

The sample size in this study is small, with only five animals per treatment group. There was high variability of behaviours between bandicoots, as reflected by the large standard errors for some analyses. Ideally, we would have included a larger sample size, however we were constrained by approvals and logistical issues. Therefore, we acknowledge our results should be interpreted carefully.

The bandicoots may have reacted to the size of the predator rather than the type of predator for the taxidermied and live-animal treatment groups. For example, it is possible that the bandicoots displayed different behaviours in the presence of a dog and cat but not a guinea pig as the dog and cat were bigger and therefore may be considered more threatening simply due to size, rather than because they are a recognised predator. However, the control taxidermied animal (a wallaby) was bigger than the taxidermied cat and no changes in behaviour were recorded for the wallaby compared to the baseline, indicating this may not be the case. This is similar to findings reported in Steindler *et al.* (2020), where model size did not appear to affect bettong avoidance behaviours. Determining how size plays a role in the recognition of predators would need further study, and may prove difficult to separate (Blumstein *et al.* 2002).

The behaviour of the predators in the live-animal treatment group may also play a role in how the bandicoots respond. Whilst the dogs used generally displayed predatory behaviours (such as stalking behaviours), the cat tended to be less interested in the bandicoots and displayed less predatory behaviours. This may have influenced how the bandicoots reacted towards the cat, and if the cat had been exhibiting more predator-like behaviours, the bandicoots may have responded differently. However, it was impossible to determine the effects of this within the confines of this study, and there is little literature on the topic. Furthermore, it is possible the bandicoots would have displayed different behaviours towards the live predators, had the predators not been restrained using leashes, harnesses and muzzles. The presence of the human handler may have also impacted the bandicoots' behaviours. To try and minimise this possibility, the human handler was present during the baseline period for that treatment group.

Whilst this study grouped animals by cue type (i.e. bandicoots were assigned either faeces, taxidermied animals, or live-animals), it would also be interesting to group animals by predator to determine if cue type influenced recognition. Although we have made observations of the differences in responses to different predator cues, the design of this study did not allow for statistical analysis between cue types as each animal received only one cue type. Future studies could expose each bandicoot to the different cue types rather than different predator types, to examine possible differences. This may provide further insight into the most appropriate cue types to test predator naivety.

Other factors to consider when testing prey naivety are the logistics associated with each cue type. Using live predators introduces a number of ethical and logistical obstacles, such as: the type of predator to be used, and whether this is possible; access to the predator, which may be influenced by the type of predator; biosecurity risk; safety and welfare for the handler, test subject, and predator; and suitability of the predator for use in such experiments, such as the ability to use restraints without causing undue stress. Taxidermied predators may replicate a live predator as closely as possible, and can be used multiple times, however access to these may be limited and require additional permits. For example, within this study we did not have access to a taxidermied dog or snake, which could have allowed more consistency across treatment groups had we been able to include these predator species (i.e. more similarities in the predators presented across all of the odour, taxidermied animal, and live animal groups).

Predator faeces is likely the easiest to procure, as faeces may be available from captive wildlife institutions, has minimal ethical restraints, and is easy to transport and store.

However, diet of the predator can also vastly influence how animals may respond to its faeces (Cox *et al.* 2010). For example, kangaroos show higher avoidance of faeces from tigers that had been fed kangaroo meat, than those that had been fed goat meat (Cox *et al.* 2010). Whilst this is important to consider in recognition studies, procuring faeces from a predator that has been fed the study species may be extremely difficult (and wasn't feasible for this experiment).

The use of wild-caught bandicoots in a captive situation allowed for the control of some environmental variability and allowed the testing of the same individuals with multiple predators. However, we acknowledge that the captive situation may have altered the bandicoots' behaviours as compared to the wild. To limit any potential of this occurring, testing for recognition of predators was conducted relatively soon after arrival.

Implications

The results of this study show that bandicoots, and likely other Australian mammal species, may benefit from assistance in learning how to recognise and better respond to predators and their cues. This could be done with the aid of predator avoidance training, which aims to train animals to recognise and respond appropriately to predators (McLean *et al.* 1994), and has shown to be successful with other mammalian species (Shier and Owings 2006; Ross *et al.* 2019; Tay *et al.* 2021). This may be particularly relevant for captive-bred, isolated, or translocated animals which may display even less antipredator behaviours than their wild counterparts (Jolly and Phillips 2020; Read *et al.* 2023). Alternatively, providing animals with refuge to make it harder for the predators to hunt may increase survival (Winnard and Coulson 2008). This could include manipulation of vegetation to increase groundcover and shelter or providing animals with alternate refuge such as nest boxes or artificial hollows (McGregor *et al.* 2014; Edwards *et al.* 2020).

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Conflicts of Interest

The authors declare no conflicts of interest.

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Data availability

The data that support this study will be shared upon reasonable request to the corresponding author.

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Tables

Bandicoot	Sex	Weight	Property type	Treatment
		(g)		
K-544	Female	900	Peri-urban	Odour
J-564	Male	1800	Rural	Odour
E-549	Male	1450	Peri-urban	Odour
M-640	Female	1100	Rural	Odour
R-303	Male	560	Peri-urban	Odour
K-545	Male	1200	Peri-urban	Live
J-562	Female	1100	Rural	Live
E-553	Male	1500	Peri-urban	Live
D-678	Male	2700	Peri-urban	Live
R-965	Male	2400	Peri-urban	Live
J-560	Female	600	Rural	Model
E-550	Male	1500	Peri-urban	Model
D-677	Male	2000	Peri-urban	Model
M-682	Female	900	Rural	Model
R-677	Male	700	Peri-urban	Model

Table 1: Details of the bandicoots tested for predator recognition and their treatment

 groups

Table 2: Partial ethogram of bandicoot behaviours used to code behavioural activity.

Behaviour	Definition
Vigilance	Investigative behaviours such as sniffing, watching, tail twitching,
	darting, and approaching food bowls cautiously (slowly and sideways).
Locomotive	Walking, hopping, or running in the enclosure, approaching food bowls
	normally.
Feeding	Bandicoot eating food, including the selection of food and chewing.
Other	Behaviours not described above, including grooming, digging, being
	stationary, or interacting with the microchip-automated nest box.
Out of view	Bandicoot was not within view of the camera, or specific behaviour could
	not be seen.

Figure captions

Figure 1: Mean (\pm SE) duration of behaviours displayed by the bandicoots (N=5) when in the presence of different predator (and control) odours, compared to the treatment baseline means represented by the dotted line (\pm SE shaded in grey).

Figure 2: Mean (\pm SE) duration of behaviours displayed by the bandicoots (N=5) when in the presence of different predator (and control) taxidermied animals, compared to the treatment baseline mean represented by the dotted line (\pm SE shaded in grey).

Figure 3: Mean (\pm SE) duration of behaviours displayed by the bandicoots (N=5) when in the presence of different live (and control) predators, compared to the treatment baseline mean represented by the dotted line (\pm SE shaded in grey).