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This research on northern quolls was carried out in accordance with protocols approved by the University of Technology Sydney Animal Care and Ethics Committee (UTS ACEC 2012-432A) and permits obtained from the Western Australia Department of Parks and Wildlife (Licences SF009187 and SF009829).

Photographs by Jonathan Webb

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Summary

The spread of the toxic cane toad *Rhinella marina* threatens populations of the endangered northern quoll *Dasyurus hallucatus*. We identified quoll populations at risk from toad invasion in the central Kimberley and explored whether free ranging quolls would consume ‘toad-aversion’ baits that induce aversions to live toads. A long-term study in Kakadu National Park showed that each generation of quolls learns to avoid toads, so one deployment of toad-aversion baits could protect quolls from toads. Encouragingly, 50% of wild quolls at Sir John Gorge, Mornington Wildlife Sanctuary (central Kimberley) consumed toad-aversion sausages. More research on captive quolls is necessary to develop long-lasting toad-aversion baits suitable for aerial deployment.

1 Focus and significance of the project

Northern Australia is currently experiencing a rapid, widespread collapse of its small mammal fauna (Woinarski et al. 2010; Woinarski et al. 2011). One species at risk of extinction is the northern quoll, *Dasyurus hallucatus*. Populations of northern quoll have rapidly gone extinct across northern Australia as cane toads invaded their range (Woinarski et al. 2014). Although quolls were declining before toads arrived, this decline was mild relative to the catastrophic impact of toads. Quolls readily attack toads but have little resistance to the toads’ toxin, and die after mouthing large toads (Covacevich & Archer 1975). If we do nothing, it is likely that the toads will eventually cause the widespread extinction of quolls in Australia’s Kimberley and Pilbara regions. Unfortunately, we cannot do anything to prevent the spread of toads through the Kimberley (Tingley et al. 2013). What we can do, however, is teach wild quolls to avoid eating cane toads (O’Donnell, Webb & Shine 2010). Populations of ‘toad smart’ quolls that avoid cane toads as prey will have a much lower risk of extinction than populations of toad naïve quolls.

Potentially, we could train wild quolls to avoid eating cane toads by deploying ‘toad-aversion’ baits (toad sausages containing a nausea inducing chemical) ahead of the toad invasion front. Quolls that consumed such baits would become ill, and would subsequently associate the smell and taste of cane toads with illness, and some individuals would ignore live cane toads (O’Donnell, Webb & Shine 2010). This process is called conditioned taste aversion (CTA), and it occurs when predators ingest novel toxic prey, become ill, and subsequently associate the smell and taste of prey with illness, and avoid consuming the prey (Garcia, Hankins & Rusiniak 1974). Provided some ‘toad smart’ female quolls survive in a toad-infested landscape, then their offspring will learn to avoid toads. This aversion is likely to be transmitted to subsequent generations via social learning (i.e. when juveniles forage with their mothers), or via CTA. For example, juvenile quolls that attack or ingest small, non-lethal metamorph toads are likely to become ill and subsequently reject toads as prey, as do smaller dasyurid predators (Webb, Pearson & Shine 2011).

The key aims of this project were to:

- Determine whether each generation of quolls learns to avoid toads as food
- Develop camera trap methods for estimating quoll population size
- Identify northern quoll populations in the central Kimberley at risk of toad invasion
- Evaluate whether wild Kimberley quolls would consume toad-aversion baits.
The research is significant because it tackles an important conservation issue; northern quolls are critically endangered in the NT, and populations in WA are vulnerable to extinction from cane toads. Our project aims to develop a method for preventing declines in northern quolls driven by the invasion of the cane toad. In halting the ongoing decline of quolls across northern Australia, we would not only be saving numerous local populations from extinction, but we will also be saving their unique genetic heritage, giving the species a greater chance of persisting through future threats (e.g. climate change). The quoll is also of cultural importance to many of the Indigenous people of northern Australia, and so conserving local quoll populations, or bringing them back to country, also allows the conservation of relevant Indigenous culture.

2 Distinctiveness of issue to this landscape

Northern quoll populations have declined across northern Australia over the last 40 years through the combined impacts of grazing, feral predators, and altered fire regimes (Braithwaite & Griffiths 1994). Cane toads are now dispersing very rapidly across northern Australia, at a rate of around 40-60 km per year (Phillips et al. 2007), and have already invaded large parts of the eastern Kimberley. At their present rate of spread, toads will have completely colonised the rest of the Kimberley within a decade (B. Phillips, personal communication 2014). Thus, we have little time left to act if we are to prevent widespread extinctions of quolls in the Kimberley.

Figure 1: A quoll stalking a cane toad.
3 Knowledge status and constraints

The broad-scale deployment of toad-aversion baits to quoll populations prior to toad invasion has the potential to reduce toad impacts on this endangered species. However, research into the application of toad-aversion baits to train wild quolls to avoid eating cane toads was not considered as an action in the national recovery plan for northern quolls because the authors suggested that ‘the likelihood of such a treatment having an effect past the initial generation of quolls is small’ (Hill & Ward 2010). Thus, long-term studies are necessary to determine whether ‘toad-smart’ quolls transmit toad-avoidance to their offspring. More importantly, we do not know if wild quolls from the central Kimberley will consume toad-aversion baits. Nor do we know if wild quolls that eat toad-aversion baits have higher survival following toad invasion compared to toad-naïve quolls.

There is also limited knowledge on the distribution of quolls in the central Kimberley or the size of extant populations. There is uncertainty surrounding the best methods for detecting quolls and estimating population sizes. The traditional method for estimating density is to live trap quolls with wire cages; however, this method is labour intensive, and is impractical for surveying large areas. Cameras provide an alternative non-invasive method for estimating density, and do not involve capture stress for target and non-target species. A recent study showed that individual quolls can be identified by unique spot patterns on their pelt (Hohnen et al. 2013). Hence, it should be possible to estimate population size in this species using remote cameras and capture-recapture analysis. At present, there is no consensus on how far apart cameras should be spaced to detect quolls and provide reliable estimates of population size.

4 Methodological approaches

4.1 Parentage analyses and monitoring of ‘toad smart’ quolls in Kakadu NP

To determine whether each generation of quolls learns to avoid eating cane toads we monitored a population of ‘toad smart’ quolls in Kakadu National Park from 2010-2014. Fifty captive reared juvenile quolls (28 males, 22 females) at the Territory Wildlife Park were trained not to eat cane toads by feeding them a small dead toad infused with the nausea inducing chemical thiabendazole (O’Donnell, Webb & Shine 2010). These ‘toad smart’ quolls were introduced to East Alligator ranger station in December 2009 and February 2010, and their long-term survival was monitored via trapping over four years. Tissue samples were taken from all individuals captured, and parentage analyses were used to determine the identity of parents of all juveniles in the population (Marshall et al. 1998; Kalinowski, Taper & Marshall 2007).
4.2 Camera trap methods to estimate quoll population size

Live trapping and camera traps were used to estimate quoll population size at Sir John Gorge (17° 31.78’S, 126° 13.08’E) at Mornington Wildlife Sanctuary in the Kimberley, Western Australia. Live trapping was done over three consecutive nights in September 2013. Small cage traps (Tomahawk Live Traps, Hazelhurst USA) baited with one tablespoon of tuna in oil were spaced approximately 30 metres apart on two transects (20 on the north transect, 10 on the south). We dusted Coopex (Bayer Environmental Science) around each cage trap to ward off ants. We checked cages at dawn and photographed each quoll, injected a microchip for identification, and recorded sex, mass and reproductive status prior to release.

We set up camera traps (Bushnell Trophy Cam, Bushnell Outdoor Products, 2012) immediately after live trapping along the same transects. We deployed 4 cameras along the south transect and 11 cameras along the north transect, with each camera spaced approximately 80 metres apart. We secured cameras to trees or rocky ledges approximately one metre from the ground facing directly downwards using Ez-Aim 2 Game Camera Mount (Slate River, LLC, Milwaukee) or a webbed strap (Fig. 2). We set cameras to high sensitivity and programmed them to take three consecutive photographs for each trigger with a delay of 10 seconds between triggers. We placed one layer of cream masking tape over the LED light of each camera to reduce the harshness of the flash, and avoid overexposure of photographs (De Bondi et al. 2010). A perforated plastic vial filled with tuna was affixed to a rock below each camera (Fig. 2). A pilot study showed that tuna was the most effective bait for detecting quolls (Austin 2014).

Figure 2: Photographs of two camera stations set up on a rock ledge (left) and in a tree (right).
Using photographs collected from the cameras, we identified individuals from spot patterns (Hohnen et al. 2013) and compiled capture histories for each individual. We estimated population size using Huggins P and C population models for closed populations in Program MARK (version 7.2). To determine how many cameras should be deployed to provide reliable estimates of population size, we ran a block bootstrapping analysis in R, using cameras along the northern transect (n = 11) with a moving block of three cameras. To determine the ideal spacing of cameras we calculated the mean number of individuals recorded from all possible combinations of cameras, alternating cameras (160 metres), every third camera (320 metres), every fourth camera (400 metres) and every sixth camera (480 metres).

4.3 Quoll surveys in the central Kimberley

Northern Quolls were surveyed across Australian Wildlife Conservancy Sanctuaries to produce baseline distribution data ahead of the arrival of cane toads. Twenty-nine sites were surveyed in 2014. At each site, six white-flash camera traps (Reconyx, PC850 Hyperfire White Flash) were set each 100 m apart in a transect following likely quoll habitat (creekline, cliffline, gorge). Cameras were positioned to face down, as described previously. We baited traps with oats, peanut butter and fish and retrieved cameras two weeks later. We identified individual quolls from images by inspecting unique arrangements of spots to generate the minimum number known to be alive for each site.

4.4 Field trials of toad-aversion baits at Mornington Wildlife Sanctuary

We made toad-aversion baits by removing the legs from freshly thawed adult cane toads that were collected by community groups during ‘toad busts’. We placed both skinned and unskinned toad legs (90 and 10% by volume respectively) into a blender to create a toad mince. We added approximately 60 milligrams of the nausea inducing chemical thiabendazole to each 15 g portion of mince. Toad meatballs consisted of 15 g portions of mince, whereas toad sausages consisted of 15 g of mince encased within a synthetic sausage skin.

Baiting trials were carried out on the southern side of Sir John Gorge in October and December 2014. Cane toads had not yet colonised this site, but could invade during early 2015. For each trial, we deployed 30 cage traps along a transect running parallel to the gorge with each trap spaced approximately 20 m apart. We placed the toad-aversion bait (either meatball or sausage) inside each trap at dusk, and checked the traps the following morning. For each trap, we recorded whether the quoll had eaten or partially eaten the bait, and recorded the microchip number, sex, mass, and reproductive status of the quoll. New quolls were injected with a microchip in the loose fur on the scruff of the neck. New baits were placed in each trap each evening, and traps were open for four consecutive nights in October, and three consecutive nights in December 2014.

Once trapping was completed, we set up nine camera traps (Reconyx, PC850 Hyperfire White Flash) spaced approximately 80 m apart along the trapping transect. Each camera was set up facing down, and was programmed to high sensitivity motion trigger as well as a time-lapse setting to take photos every 30 minutes. A toad sausage or toad meatball was placed underneath each camera, and was surrounded by a ring of Coopex to discourage ants from eating the bait. Cameras were brought in four days after deployment, and any quolls taking or investigating the toad-aversion baits were identified from their spot patterns.
5 Lessons learnt for this landscape

5.1 Parentage analyses and monitoring of ‘toad smart’ quolls in Kakadu NP – each generation of quolls learns to avoid toads

Parentage analyses of juvenile quolls captured at East Alligator revealed that each generation of quolls learns to avoid eating cane toads (Cremona et al. 2015). For example, in May 2011, there were five breeding females on the study site: two trained reintroduced females, one wild female, the offspring of a wild female, and the offspring of a trained female. These females reproduced, and in 2012 we captured 14 juveniles: 2 could not be assigned a mother, 4 were offspring of a trained reintroduced female breeding in her second year and 4 were second generation offspring of a reintroduced female (Cremona et al., 2015). Thus, some quolls learn to avoid eating cane toads. Juvenile quolls spend extended periods with their mothers (Fig. 3) and may have learnt not to eat toads by watching their mothers sniff and reject toads, or they may have ingested small non-lethal sized toads that induce nausea and long-lasting aversion to live toads in other dasyurids (Webb et al. 2008; Webb, Pearson & Shine 2011).

Figure 3: A quoll and her young.

Radio-telemetry revealed that predation by camp dogs and dingoes were the major sources of mortality for quolls on the study site. Our study site was burnt annually during the late dry season, and there was little cover available to quolls in December when young quolls begin foraging. Previous studies have shown that quolls are most at risk from dingo predation in recently burnt areas (Oakwood 2000). Population viability modeling suggests that high mortality from predation, coupled with frequent late dry season burning, is preventing the quoll populations in Kakadu National Park from recovering after toads invaded (Cremona, Crowther & Webb 2015).
5.2 Camera trap methods to estimate quoll population size

Camera traps provided good estimates of quoll population size. Live trapping yielded an estimate of 14 animals, while cameras yielded an estimate of 15 animals. Ten of the 11 cameras along the northern bank of Sir John Gorge recorded 971 photographs of quolls, of which 938 (96.6%) could be used to identify individuals. In total, we identified 14 individuals (6 females, 7 males and one unknown). Comparing photographs taken during live trapping to photos from cameras, we determined that all live captured individuals were captured on remote cameras. An additional individual was captured on camera but not during live trapping. Bootstrapping revealed that the number of individuals recorded on camera decreased as cameras were more widely spaced (Fig. 4). At Sir John Gorge, spacing of cameras 80 m apart provided reliable estimates of quoll population size (Fig. 4).

![Figure 4: Mean number of individual quolls detected at different camera spacing (± standard errors).](image)

5.3 Distribution of quolls in the central Kimberley

Twenty-nine sites were surveyed in 2014 (Fig. 5). Additional data collected between 2011 and 2014 is included in the map for sites in the north-western parts of Artesian Range where quolls are common. Quolls were present at 9 of the 25 sites surveyed on Mornington, Marion Downs and Tableland sanctuaries, all of which were within a discrete area of the King Leopold Ranges in the south-east of Mornington (Fig. 6). At these sites, the minimum number of quolls detected on cameras ranged between 1 and 12 (Table 1). These small populations are vulnerable to extinction from toad invasion and environmental stochasticity.
Figure 5: Distribution of the northern quoll on AWC sanctuaries in the Kimberley. The current location (September 2014) and direction of spread of the cane toad invasion front is shown in green. The next nearest quoll population to the northwest is shown in the orange ellipse; there are no quoll populations to the northeast of the Mornington population.

Table 1. Northern quoll populations on Mornington and the neighbouring crown land with minimum number known to be alive from most recent survey data, data source and watercourse.

<table>
<thead>
<tr>
<th>Site</th>
<th>Minimum number known to be alive</th>
<th>Data source</th>
<th>Watercourse</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sir John Gorge</td>
<td>12</td>
<td>Trapping 3 x annually</td>
<td>Fitzroy River</td>
</tr>
<tr>
<td>Tin Can Gully</td>
<td>9</td>
<td>Camera traps 2014</td>
<td>Fitzroy River</td>
</tr>
<tr>
<td>Rose's Pool</td>
<td>8</td>
<td>Camera traps 2014</td>
<td>Spider Creek</td>
</tr>
<tr>
<td>Cowendyne South</td>
<td>9</td>
<td>Camera traps 2014</td>
<td>Cowendyne Creek</td>
</tr>
<tr>
<td>Sir John Gorge Upper</td>
<td>3</td>
<td>Camera traps 2014</td>
<td>Fitzroy River</td>
</tr>
<tr>
<td>Cliftoniana Gully</td>
<td>10</td>
<td>Camera traps 2014</td>
<td>Fitzroy River</td>
</tr>
<tr>
<td>Collis Creek</td>
<td>1</td>
<td>Camera traps 2014</td>
<td>Fitzroy tributary</td>
</tr>
<tr>
<td>Cowendyne North</td>
<td>-</td>
<td>Trapping 2011</td>
<td>Cowendyne Creek</td>
</tr>
<tr>
<td>Narrie West</td>
<td>4</td>
<td>Camera traps 2014</td>
<td>Fitzroy tributary</td>
</tr>
<tr>
<td>Narrie Range Sir John</td>
<td>3</td>
<td>Camera traps 2014</td>
<td>Fitzroy River</td>
</tr>
<tr>
<td>Slingshot Gap</td>
<td>-</td>
<td>Trapping 2011</td>
<td>Fitzroy tributary</td>
</tr>
</tbody>
</table>
5.4 Field trials of toad-aversion baits at Mornington Wildlife Sanctuary

In October, we trapped five quolls in traps baited with toad-aversion baits, and five quolls were identified on camera taking baits. In December, we trapped two quolls; one new female (not trapped in October) consumed the bait, while one male trapped in October did not consume the bait. Quolls investigated baits at five of the cameras, sometimes rubbing their bellies over the baits. No additional quolls to those that inspected baits in October were recorded. One individual investigated baits nine times at four different cameras but didn’t eat them. This individual previously took two toad-aversion baits in the October camera trap trial. A second quoll investigated baits seven times at five cameras but did not eat them. Again, this female had previously eaten a bait in the October trial. Two quolls from the October trial that hadn’t taken baits investigated baits in December.
6 National implications of lessons learnt

Our research in Kakadu National Park demonstrated that each generation of quoll learns to avoid toads as prey. Thus, toad aversion baiting would only need to be done once to protect quoll populations from cane toads. Thereafter, each generation of quolls can learn to avoid eating cane toads, perhaps via social learning, or via ingestion of small non-lethal toads that induce aversions to live toads.

Camera traps, in conjunction with identification of individuals via spot patterns, can provide reliable estimates of quoll population size in the central Kimberley. Using this methodology, we identified another eight quoll populations near Mornington Wildlife Sanctuary, all of which are small, and thus, vulnerable to extinction (Table 1).

Six of 12 quolls (50%) at Sir John Gorge ingested toad-aversion baits. The major problem that we observed was the toad aversion baits went rancid overnight, which presumably made them less palatable to quolls. Hence, we either need to develop long-lasting toad-aversion baits that are more palatable to quolls, or we need to bring quolls into captivity prior to toad invasion to prevent population extinctions. Our research in Kakadu National Park shows that trained ‘toad-smart’ quolls can be reintroduced following toad invasion, so the latter option should be feasible.

7 Problems addressing the focus and how to overcome these

7.1 Cane toad invasion slower than anticipated

The invasion of cane toads was slower than anticipated, and cane toads did not invade the study sites in 2014. Consequently, we were unable to experimentally test whether wild quolls that consumed toad-aversion sausages had higher survival than toad naïve individuals following toad invasion. This problem will be overcome by deploying toad-aversion baits to experimental sites in 2015, and monitoring the subsequent survival of radio-collared quolls at control and experimental sites before and after cane toads invade the study sites.

7.2 Toad-aversion bait shelf life

We did not impregnate toad-aversion baits with salts, preservatives or fats, and consequently the toad-aversion baits rapidly desiccated and went rancid overnight during the December baiting trials when day time temperatures exceeded 45 °C. Future studies are necessary to develop long-lasting toad-aversion baits suitable for aerial deployment. A captive colony of quolls is crucial for testing the efficacy of the toad-aversion baits for inducing aversions to live toads. Currently, no captive quolls are available to do this research.
8 Towards implementation

Toads are expected to arrive at Mornington Wildlife Sanctuary either in February-March 2015, or early 2016. Our plan is to field test whether toad-aversion baiting can reduce the impacts of cane toads on quoll populations in the central Kimberley. We will deploy toad-aversion baits in the field, and monitor the fate of experimental (toad-aversion baits deployed) and control (no baits deployed) populations via camera trapping. A more intensive study will be done at Sir John Gorge. At this site, toad-aversion baits will be placed in cage traps, and the fate of quolls that consume baits (trained quolls) or do not eat baits (untrained quolls) will be monitored via radio-telemetry before and after toad invasion.

9 Looking ahead – future needs

Cane toads are rapidly spreading across the Kimberley, and will likely cause quolls in that region to go locally extinct. There are two strategies that could be used to prevent quoll populations from going extinct:

1. Develop long-life toad-aversion baits suitable for aerial deployment, and deploy these baits ahead of the toad invasion front.

We need to develop toad aversion baits with a long shelf life suitable for aerial deployment. The baits we tested in this study rapidly desiccate and go rancid overnight, and thus quickly become unpalatable to quolls. The incorporation of toad fats and tasteless, odourless preservatives into the next generation of baits could help to solve this problem. A captive colony of quolls will be necessary to test whether this next generation of toad-aversion baits can induce an aversion to live toads in quolls. The Territory Wildlife Park has purpose built quoll enclosures available to house quolls and TWP personnel have considerable expertise in maintaining and breeding quolls. Wild quolls could be obtained from Pobassoo and Astell Islands off Gove. These island quolls are derived from wild stock obtained from the Darwin and Kakadu regions, and because they are toad-naïve, they are ideal subjects for testing the efficacy of next generation toad-aversion baits.

After development and testing on captive quolls, next generation toad-aversion baits could be deployed via helicopter ahead of the toad invasion front. We have identified several quoll populations that could serve as control populations (no baiting) and experimental populations (baiting) near Mornington Wildlife Sanctuary (Table 1). These populations could be monitored via camera trapping before, during, and after toad invasion to test whether toad-aversion baiting prevents quoll populations from going locally extinct.

2. Capture wild quolls before toads invade and maintain the quolls inside toad-proof enclosures at Mornington Wildlife Sanctuary

Bringing wild quolls into captivity before the toads invade would prevent local extinctions from occurring. Once cane toads have invaded, the offspring of wild quolls could be trained not to eat cane toads, by feeding them a small dead toad laced with thiabendazole (O’Donnell, Webb & Shine 2010). These ‘toad smart’ quolls could then be reintroduced to the wild after the toads have invaded (Fig. 7). The research at Kakadu National Park suggests that some of these quolls are likely to survive and reproduce, and their offspring are also likely to learn not to eat cane toads.
Figure 7: A quoll is released after being microchipped and having its sex, mass and reproductive status recorded.

Figure 8: Cane toad *Rhinella marina*. 

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