

Reducing the primary exposure risk of Henderson crakes (*Zapornia atra*) during aerial broadcast eradication by selecting appropriate bait colour

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Abstract

Context. Operations to eradicate non-native invasive predators from islands frequently put native species at risk of consuming harmful substances, such as poison bait. The incorporation of certain colours in poison-bait pellets may reduce the risk of bait consumption and, therefore, non-target mortality. Previous work indicated that birds generally avoid blue or green colours; however, there is substantial inter-specific variation in this preference, and more experimental work on species of conservation concern is needed.

Aims. We tested whether a globally threatened island endemic, the Henderson crake (*Zapornia atra*), which suffered substantial mortality during a rat-eradication attempt on Henderson Island in 2011, would consume fewer blue than green pellets, which were used during the previous eradication attempt.

Methods. We held 22 Henderson crakes in captivity and provided them with either blue or green non-toxic pellets for 5 days in June and July 2015. We measured consumption and used linear mixed models to evaluate whether bait colour influenced consumption.

Key results. Henderson crakes did not consume any dry pellets, and all trials were conducted with wet bait pellets. We found slightly lower consumption of blue pellets than green pellets, and substantial variation among individuals. Females ($n = 17$) consumed 24% less blue than green bait, whereas males ($n = 5$) consumed 77% less blue than green bait.

Conclusion. Henderson crakes are unlikely to consume dry pellets, and will likely consume fewer blue than green bait pellets.

Implications. We recommend that any future rat eradication on Henderson Island considers using blue rather than green baits and targets dry weather to reduce the risk of Henderson crakes consuming toxic rodenticide bait pellets.

Additional keywords: Henderson Island, non-target mortality, poisonous baits, primary poisoning, rail.

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Introduction

Non-native predators pose a significant threat to many bird populations, especially on offshore islands. Eradicating invasive mammal species, such as rats (*Rattus* spp.), from islands has become an increasingly common conservation approach to protect native biodiversity (Towns and Broome 2003; Veitch *et al.* 2011; Simberloff *et al.* 2013). A critical consideration in planning an eradication is the extent to which the operation may put native species on the island at risk, particularly species endemic to the island (Thomas and Taylor 2002; Zavaleta 2002; Oppel *et al.* 2011).

Rats are cosmopolitan predators of birds and have caused the decline or extinction of many island populations (Atkinson

1985; Jones *et al.* 2008). Rats have been eradicated from >500 islands worldwide, and standard guidelines are available for how to plan for and implement rat eradication on islands (Howald *et al.* 2007; Keitt *et al.* 2015). The most successful and widely applied technique to eradicate rats from islands >100 ha is aerial broadcast of toxic bait pellets at a sufficient density to ensure all rats on the island have access to and consume sufficient toxic bait. This technique also puts all animals in the natural environment inhabited by rats at risk of consuming toxic bait. Many species are either physiologically unaffected by the chosen toxin, or behaviourally unlikely to consume grain-based pellets from the ground. However, some species not targeted by an eradication operation can suffer substantial mortality through

direct consumption of bait (primary poisoning) or scavenging on poisoned individuals (secondary poisoning; Spurr and Powlesland 1997; Hoare and Hare 2006). Minimising this non-target mortality is generally important for an eradication operation to achieve its desired objective, namely the conservation of native species and the restoration of island ecosystems.

One way to reduce the attractiveness of bait pellets to non-target species is to use a colour that is naturally less attractive for the non-target species, while being equally likely to be consumed by rats (Hartley *et al.* 1999, 2000; Weser and Ross 2013). Previous work on colour preferences in birds generally found that colours such as red and yellow are preferred over colours such as blue, green or brown (Caithness and Williams 1971; Brunner and Coman 1983; McPherson 1988). However, which of the latter colours is less preferred appears to vary considerably among species. For example, weka (*Gallirallus australis*) and New Zealand robins (*Petroica australis*) avoided blue bait the most (Hartley *et al.* 1999, 2000), but Steller's jays (*Cyanocitta stelleri*) and kea (*Nestor notabilis*) avoided green more than blue (Slaby and Slaby 1977; Weser and Ross 2013). There is, therefore, a need to identify more species-specific colour preferences to reduce the potential risk of non-target mortality during aerial poison applications (Clapperton *et al.* 2014; Veltman *et al.* 2014).

Here, we examined whether a species of flightless rail, the Henderson crane (*Zapornia atra*), consumed more bait of either blue or green colour. Henderson cranes are endemic to Henderson Island in the Pitcairn Islands of the South Pacific, a very remote and largely undisturbed raised coral atoll not currently inhabited by humans. Pacific rats (*R. exulans*), which were introduced by Polynesian settlers, negatively affect seabird populations on this island (Brooke *et al.* 2010), and an aerial broadcast rat eradication using green grain-based brodifacoum pellets was conducted in 2011 (Torr and Brown 2012). *Porzana* and related cranes are known to be vulnerable to aerial poison-bait applications (Spurr and Powlesland 1997). Mortality of cranes was, therefore, anticipated during the 2011 eradication and mitigated by keeping a sufficiently large 'rescue' population in temporary captivity during the eradication attempt (Brooke *et al.* 2011). This captive population ultimately facilitated the rapid recovery of the wild Henderson crane population within 2 years after the eradication (S. Oppel and A. Bond, unpubl. data). Nonetheless, large mortality of wild Henderson cranes occurred during the eradication operation in 2011, inferred from the confirmed death of birds monitored with radio-transmitters after the bait drop (Brooke *et al.* 2011). The timing of mortality was consistent with birds consuming wet green bait pellets, because secondary poisoning was experimentally determined to be unlikely (Brooke *et al.* 2013). Because the eradication was unsuccessful and may be repeated in the future using a similar approach, information is required on whether Henderson cranes might consume fewer bait pellets of a different colour. We therefore captured Henderson cranes and tested whether controlled intake of blue pellets would be lower than that of green pellets. This experiment, therefore, provides some guidance on how the non-target mortality of a globally threatened endemic bird species during an aerial broadcast eradication might be reduced, and may be relevant for other

flightless bird species at risk during rodent-eradication operations.

Materials and methods

Study area and climate

The study was conducted on Henderson Island (24°20'S, 128°20'W) in June and July 2015. During this time period, the temperature in the environment where the cranes were held in captivity was 12–30°C, with a mean of 21.2°C and the mean relative humidity was 79%, ranging from 48% to 100%.

Crane capture and captive husbandry

Henderson cranes were captured in the natural forest on Henderson Island, within 1000 m of the research station on North Beach. Cranes were captured using mist nets ($n=22$), spring-based traps with a small water bowl as lure ($n=11$) and a hand net ($n=1$). The sex of captured cranes was determined by using the colouration of their bill and legs (Jones *et al.* 1995). Captured cranes were transferred to individual aviaries situated under shady vegetation next to the research station, along the beach. Individual aviaries were $2 \times 3 \times 1$ m in size and contained sufficient vegetation for birds to hide from view and shelter from rain. Cranes that did not investigate and consume food provided in food bowls and therefore did not acclimatise to captivity ($n=6$ males and 4 females) were released back into their original territory within 3–5 days after capture. Two birds died in captivity before trial initiation. On completion of the feeding experiment, all remaining rails ($n=22$) were released back into their original territories.

Birds were provided with water *ad libitum* and food designed to support the high protein requirements of avian insectivores (Wombaroo Insectivore Rearing Mix, Wombaroo Food Products, Adelaide, SA, Australia) for 7 days before the provisioning of experimental bait pellets. The Wombaroo diet consisted of a brown paste that was presented in a creamy state in a food bowl and did not resemble the experimental pellet diet in either colour, size, shape or consistency. Because handling and weighing of birds was stressful, birds were weighed only at capture and release at the end of the feeding trial to provide information on whether the provided food was adequate for birds to maintain roughly stable mass. Food was provided at 0630 hours (local time, UTC-8) every morning, and any remaining food was collected at 1730 hours every evening to minimise attracting rats and crabs to aviaries at night. This research was approved by the Government of the Pitcairn Islands and the RSPB Ethical Advisory Committee (Protocol EAC 2015/01).

Feeding trial

Once the birds had habituated to the captive environment, individuals were assigned randomly to receive either green or blue, but otherwise identical, pellets as an experimental diet. The pellets were surface coated with blue and green dye, and did not lose their colouration when soaked in water for several hours. The green dye was a composite of tartrazine powder (with the Chemical Abstracts Service (CAS) number 1934-21-0), Brilliant Blue powder (CAS number 3844-45-9) and sodium sulfate

(CAS number 7757-82-6), and the blue dye was Hexacol Indigo Carmine Supra Blue R2613 (CAS number 860-22-0).

We provided rails with only one pellet colour, because during an eradication operation the rails would not be exposed to different bait colours, and a direct choice test was not appropriate. Our test was therefore designed to assess the consumption of a novel food type, rather than test whether the crakes chose one colour over another when both were available.

The experimental diet was provided every other day for a 10-day trial period. Birds were presented with 10 wet non-toxic bait pellets (mean dry mass of each pellet 1.8 g; Pestoff™, Whanganui, New Zealand) of a single colour that had been soaked in water for 10 h, simulating pellets that had been soaked by rain during an eradication operation. Initial trials with dry bait pellets indicated that crakes were unable to consume dry pellets (see Results), and no dry pellets were provided after the first week, to increase the relevant sample size of wet pellets. The experimental pellet diet was provided at 0630 hours, and replaced with food *ad libitum* after 6 h. The mass of provided and retrieved pellets was weighed with an electronic balance accurate to 0.1 g. Many crakes spread pellets in the aviary without consuming any pellets. We attempted to retrieve pellets from the leaf litter in each aviary, and noted whether birds had actually consumed pellets (evident by peck marks on the soft pellets) or simply dispersed pellets in the cage without consuming them.

Analysis

To determine whether either of the two bait colours was consumed more than the other, we first calculated the amount of pellets consumed by subtracting the mass retrieved from the mass provided for each individual bird and experimental day. Days on which heavy rain flooded food bowls were excluded. Instances where the rails dispersed pellets in their respective aviary without any evidence of consumption were set to 0, to avoid erroneous interpretation of pellet consumption as a result of pellets being lost in aviaries.

We used generalised linear mixed models (GLMM) to assess the effect of bait pellet colour on crake pellet consumption. We accounted for serial autocorrelation at the individual level by including individual crake identity as a random intercept in each model (Bolker *et al.* 2009). Because we caught an uneven number of males and females, we used a multi-model inference approach (Burnham and Anderson 2002) to evaluate the effect of bait colour on consumption of pellets. We fitted five plausible models that assumed that (1) pellet consumption was independent of bait colour and sex, (2) consumption varied by bait colour, (3) consumption varied by sex, (4) consumption varied by sex and bait colour (additive fixed effects), and that (5) consumption differed between sexes and bait colours (interactive effect). Because pellet consumption may have increased over time as birds became accustomed to pellets, we included 'trial day' as a fixed effect in all models. All models included the same random effect, and we used the second-order Akaike's information criterion corrected for small sample sizes (AIC_c) to assess the support for each of the five models (Burnham and Anderson 2002). We fitted all models by the Laplace approximation in R 3.1.3

(R Development Core Team 2014) with the package 'lme4' (Bates *et al.* 2014). We present mean parameter estimates and standard errors for the most parsimonious model, determined by the lowest AIC_c value.

Results

We captured a total of 34 crakes, of which 22 (17 female, 5 male) habituated to captivity and participated in the feeding trial. We assigned nine females and two males to the group receiving blue pellets, and eight females and three males to the group receiving green pellets. No bird consumed any dry pellets during 15 experimental-trial feeding days with eight birds. Excluding days where dry pellets were provided or where the food bowls were flooded by rain, we obtained data from 90 trial feeding days from 22 individuals. During the 6 h of experimental feeding, birds generally consumed between 0 and 25.8 g (0–98%) of the provided wet bait pellets (Tables S1, S2, available as Supplementary material to this paper). Over the 17 days of captivity, females gained, on average, 12.5% body mass (difference between capture and release 8.8 ± 6.1 g, paired Student's *t*-test, $t_{13} = 5.41$, $P < 0.001$), whereas males maintained their body mass (difference -0.3 ± 13.7 g, $t_3 = -0.081$, $P = 0.94$).

Of the five models we evaluated to explain bait consumption, the model specifying that pellet consumption differed between males and females and by bait colour received overwhelming support from the data (Table 1). The additive model including sex and bait colour also received some support, and both models indicated slightly higher consumption of green pellets than blue pellets by both sexes (Table 2). Female crakes

Table 1. Model-selection table evaluating five hypotheses regarding variation in consumption of bait pellets by Henderson crakes during a captive-feeding trial in June and July 2015

See text for description of models. AIC_c , the second-order Akaike's information criterion adjusted for small sample sizes; ΔAIC_c , the difference between each model and the most parsimonious model; and ωAIC_c , the weight of evidence for each model; k , the number of parameters in each model

Model	k	AIC_c	ΔAIC_c	ωAIC_c
Sex \times colour	7	580.45	0.00	0.78
Sex + colour	6	583.39	2.94	0.18
Sex	5	586.67	6.22	0.03
Colour	5	589.97	9.52	0.01
Null	4	592.60	12.15	0.00

Table 2. Estimated bait-pellet consumption by Henderson crakes during a captive-feeding trial in June and July 2015

Daily consumption of bait pellets (in g) was estimated with the most parsimonious model for the midpoint of the trial, including sex and bait colour as interactive fixed effects (Table 1)

Bait colour	Sex	Mean estimate (g)	Standard error
Blue	Female	8.71	4.79
Green	Female	11.57	4.83
Blue	Male	1.54	5.79
Green	Male	6.15	5.41

consumed, on average, more bait pellets than did males, but the difference between blue- and green-pellet consumption was smaller for females than for males (Table 2). The model also indicated that pellet consumption increased by 0.63 g (± 0.20 s.e.) per trial day.

Discussion

Henderson crakes did not consume any dry pellets in our experimental feeding trial, and may be vulnerable to primary poisoning during an eradication operation only when substantial rain renders the pellets soft and palatable. We found slightly lower consumption of blue bait pellets than green pellets, and substantial individual variation (Table S1), with females generally consuming more bait pellets than did males. Females also appeared to differentiate less between blue and green pellets than did males, but this effect may be a consequence of the small sample size of males. We therefore recommend that any future rat-eradication operation on Henderson Island considers using blue bait pellets instead of green pellets and continues to aim for dry weather conditions to reduce the primary exposure of Henderson crakes to rodenticide contained in bait pellets. However, although bait consumption may be reduced by using a different colour, most birds in our trial consumed at least some wet pellets. Given the toxicity of pellets used during eradication operations, it is likely that there will be some mortality associated with any aerial bait application, and our results do not suggest that accidental mortality of Henderson crakes could be completely eliminated if an aerial brodifacoum-based eradication operation is repeated.

Our results are consistent with research on other bird species that also found a lower preference for blue- than for green-coloured food items (Hartley *et al.* 1999; Hartley *et al.* 2000; Clapperton *et al.* 2012), but whether these differences are caused by visual attractiveness or by palatability of different colours is unknown. However, bait colouration may not prevent bait consumption by inquisitive birds completely unless a stronger deterrent is included in the bait (Hartley *et al.* 2000; Weser and Ross 2013). In addition, aviary trials may not properly reflect food-choice preferences in the wild (Esther *et al.* 2013), and some uncertainty remains on how Henderson crakes would react to encountering blue pellets in the wild. Henderson crakes are fairly inquisitive birds with a broad dietary range (Jones *et al.* 1995), and readily consumed pink rhodamine-dyed non-toxic pellets during rat-consumption trials in 2009 (M. Brooke, pers. comm.). The consumption of green bait pellets in 2011 was speculated to have been a consequence of extremely wet weather rendering the bait pellets soft, palatable and very similar in size and colouration to an abundant sphingid moth caterpillar (*Gnathothlibus erotus*) readily consumed by birds in captivity in 2011 (G. Harrison, pers. obs.). Although blue rather than green colour may reduce the risk of primary exposure of Henderson crakes during an aerial broadcast eradication, additional bird-repelling ingredients such as anthraquinone may further reduce the risk to native birds (Clapperton *et al.* 2012, 2014; Crowell *et al.* 2016), but would have to be weighed against their efficacy for poisoning rats (Cowan *et al.* 2015). Our experiment also indicated that

Henderson crakes are unlikely to consume dry pellets, and aiming at dry weather conditions or developing more water-resistant bait pellets would likely substantially reduce the risk of bait consumption by Henderson crakes.

Other native species on Henderson Island may have different colour preferences and may be at higher risk from blue than green bait. In particular, the Henderson fruit dove (*Ptilinopus insularis*), a native frugivore, may be more attracted to blue rather than green colours (Willson *et al.* 1990). However, fruit doves consume several green fruits (Brooke and Jones 1995), and frugivore colour preferences often match the frequency of fruit available in the environment (Duan *et al.* 2014). Although there are blue fruits on Henderson (*Dianella intermedia*; Florence *et al.* 1995), they appear to be very rare and do not feature prominently in the diet of fruit doves (Brooke and Jones 1995). We therefore consider it unlikely that other native birds would find blue bait pellets more attractive than green pellets.

A modification of the bait colour used in future rodent-eradication attempts on Henderson Island requires further experiments to test whether bait acceptance and uptake rates of rodents are lower for blue bait (Clapperton *et al.* 2015), so as to avoid sacrificing efficacy for minimising risks to non-target species. Rats have comparatively poor colour vision, with eye receptors most responsive to blue-green light, but they are also sensitive to UV light (Jacobs *et al.* 2001; Burn 2008). Although current evidence suggests that rats could detect blue and green bait equally, bait acceptance may be affected by UV light or olfactory or taste differences and we therefore recommend bait-acceptance trials with blue bait before any future eradication attempt.

In summary, blue rather than green bait should be considered for future eradication attempts on Henderson Island, so as to reduce the primary exposure risk of endemic Henderson crakes to rodenticide contained in pellets.

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