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Research Article

Relative acceptance of brodifacoum pellets and soft bait sachets by Polynesian rats (*Rattus exulans*) on Wake Atoll

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Abstract

Removing invasive rats from island ecosystems using rodenticides has proven conservation benefits and is an important management tool for conserving and restoring island ecosystems. However, rodenticide-based eradications can fail if not all rats consume enough bait to result in lethal toxicosis. A recent post-operational review of a failed attempt to eradicate rats from Wake Atoll suggested that some individuals may not have ingested a lethal dose of rodenticide due to potential dietary and/or sensory preferences developed via regular access to anthropogenic food sources. These food sources may be higher in fats and oils, possessing different sensory properties (e.g., softer, chewier, etc.) than the harder pellet formulation of the rodenticide Brodifacoum 25W Conservation (B-25W) used in the eradication attempt. To test this theory, we captured rats from two areas on Wake Island where they may have regular access to human food sources, as well as an uninhabited part of island where rats presumably have less access to human-based food sources and therefore are less likely to be preconditioned for these food types. We subjected them to a head-to-head two-choice bait selection trial between a "soft" sachet formulation of a brodifacoum-based bait, FINAL Soft Bait with Lumitrack® (FINAL), versus the harder pellet formulation of B-25W. Regardless of which habitat rats were captured in, rats overwhelmingly preferred the pellet formulation. No rats in the head-to-head trail consumed any of the FINAL bait, and 100% of the rats that consumed B-25W died. Of the rats in a separate no-choice trail of just FINAL bait, 5 failed to eat any bait; of the rats that did consume some of the FINAL bait, 80% died. Our results demonstrate that Polynesian rats on Wake Atoll do not prefer this soft formulation of brodifacoum-based rodenticide bait. Our results suggest that baiting strategies in the inhabited regions of the atoll, for a proposed eradication attempt, should continue to focus on utilizing traditional pellet formulations. While these results are unequivocal in our test case, we suggest caution in making inference to other situations where dietary preferences of local rodent populations may differ, and local environmental conditions may make other baiting choices more appropriate and efficacious.

Key words: bait trials, diet preferences, island rodent eradication, rodenticide, sensory preferences, two-choice trials

Introduction

Removing invasive rats (*Rattus* spp.) from islands is an effective technique to conserve unique and endangered island species and restore island ecosystems (St Clair et al. 2011; Towns et al. 2013; Jones et al. 2016). This is usually accomplished by employing some combination of overlapping treatments of bait stations, hand sowing, and aerial broadcasting cereal-based rodenticide baits, typically containing the second-generation anticoagulant brodifacoum (Howald et al. 2007). The success of any rat eradication project relies on distributing sufficient toxic bait to every rat territory to ensure that every individual has access to enough bait to ingest a lethal dose (Broome et al. 2017). Ensuring that there are no gaps in bait coverage is clearly an important factor in successful rat eradications, but equally important is ensuring that the bait being deployed is not only palatable to the target population but is also efficacious. It is common practice to conduct food trials prior to most rat eradications to ensure that the bait used in the eradication is both palatable and efficacious. On islands with human inhabitance, there is an additional concern that rats inhabiting commensal areas may develop different food preferences than rats that only have access to naturally occurring food items. If so, bait palatability may differ among rats from different areas of an island, necessitating the identification of alternative rodenticides that appeal to the dietary and sensory preferences of rats in commensal areas or that have regular access to anthropogenic food waste.

Previous studies have documented that rats consuming a high-fat diet during developmental growth phases are known to have an increased preference for high-fat foods (reviewed in Warwick and Schiffman 1992) and laboratory rats are known to prefer high-fat and/or high-sugar foods (Sclafani 2001). Preferences for fatty food items can also develop during periods of food restriction (Lucas and Sclafani 1989). Additionally, high-fat food items differ in palatability and possess different sensory properties (i.e., softer, chewier) compared to diets consisting of other food items (Warwick and Schiffman 1992). Thus, it is possible that human foods and associated food waste can be substantially more attractive, or equally so, to rats than hard cereal-based rodenticide bait pellets typically used in island rat eradications, presumably because they are higher in fat and/or sugar content than rodenticide bait products.

In 2012, the Pacific Air Forces (PACAF), 611 Civil Engineering Squadron (611 CES), in conjunction with the United State Fish and Wildlife (USFWS) and Island Conservation (IC), attempted to eradicate both the Asian house rat (*Rattus tanezumi* Temminck, 1844) and Polynesian rat (*R. exulans* Peale, 1848) from all three islands comprising Wake Atoll. They were successful in removing all Asian house rats from the entire atoll, but not in removing all Polynesian rats from Wake Island, the only inhabited island in the atoll. At the time, this eradication operation was widely considered one of the most complex eradication projects to have been attempted, and no single factor was likely responsible for the failure to eradicate both species (Brown et al. 2013; Griffiths et al. 2014). Post-action reviews of the project identified bait preference/aversion issues coupled with availability of alternative naturally occurring or commensal foods as a potential factor in the failure to eradicate all rats from Wake Island (Brown et al. 2013; Hanson et al. 2019). Rats on Wake Island had potential access to anthropogenic food sources associated with multiple dining areas in commensal environs of the island (e.g., cafeteria, contractor apartments and residential areas) as well as in a solid waste aggregating area (SWAA) where food wastes were stored prior to incineration and accumulated operational detritus provided ample nesting opportunity. As part of the planning for a proposed future eradication attempt, we sought to address the concerns raised in these reviews regarding preconditioned dietary preference of rats inhabiting Wake Island.

Despite attempts to limit the availability of human-based food items and food waste to rats on Wake Island during the 2012 eradication, the presence of multiple dining areas in the commensal environs of Wake Island and waste storage at the SWAA presented potential opportunities for rats to encounter human foods and food waste. Thus, it has been hypothesized that individual rats utilizing these areas may have developed sensory and dietary preferences for these types of food, potentially making rats less likely to consume the lower-fat, hard, high-carbohydrate pellet formulations of baits required for aerial broadcasting (but see Samaniego et al. 2020).

Brodifacoum-25W Conservation pellets (B-25W; 25 ppm brodifacoum; Bell Laboratories, Madison, WI) is the bait product that was used in the 2012 eradication and likely to be used in a proposed future attempt. The only previous bait palatability study conducted on Wake Island confirmed that Polynesian rats found B-25W highly palatable and preferred it to naturally occurring food sources such as ironwood cones, nutsedge, noni and tournefortia fruits, and hermit crabs (Shiels et al. 2015). However, none of the items used in that study represented high fat/sugar items found in human foods and food waste. In addition, the test subjects for that study were all captured at the SWAA; while informative, the results do not indicate how rats in the other commensal areas and natural areas of Wake Island are likely to interact with different formulations of brodifacoum baits. FINAL Soft Bait with Lumitrack® (FINAL; 25 ppm brodifacoum; Bell Laboratories, Madison, WI) is another brodifacoum-based anticoagulant rodenticide that was developed specifically to appeal to rodents that were preconditioned to very fatty or oily foods (C. Riekena, Bell Labs, *pers. comm.*). FINAL baits are comprised of 15 g of bait product delivered in thin sealed plastic envelopes, or “sachets.” Being a “soft” bait, this formulation might also appeal to sensory preferences for soft food. As such, FINAL represents a bait that rats preconditioned to these properties could find attractive, making it an ideal candidate for use as an alternative bait in commensal areas where aerial baiting may be restricted. Confirming whether such dietary preferences exist or not and whether this bait is efficacious addresses an important knowledge gap in preparation for a proposed subsequent attempt to eradicate Polynesian rats from Wake Atoll and may be important wherever permanent human settlements provide conditions that might promote such preferences and application techniques other than sowing of hard pellets are preferred.

Here we present the results of a food trial designed to test if there are dietary and/or sensory food preferences among rats inhabiting commensal, waste storage, and natural areas on Wake Island and if those preferences influence efficacy of either soft or hard bait options. Because we could not find any published results on field efficacy of FINAL and it has not been used previously on Wake Island, we also ran a separate no-choice trial to assess efficacy. This information is being used to inform decisions regarding alternate baiting strategies for a proposed future eradication attempt on Wake Atoll.

Materials and methods

Study site

Located in the central Pacific (2,416 km east of Guam and 3,698 km west of Hawaii), Wake Atoll is a low-lying atoll (max elevation < 7 m) comprised of 3 islands (Wake Island, Peale Island, and Wilkes Island, 7.1 km^2 total) that is managed by the US Air Force (USAF). Both Peale Island and Wilkes Island do not have any permanent human inhabitants, but the USAF and supporting contractor staff make up a persistent community that fluctuates between < 100 to < 500 people on the main island of Wake.

Sample collection and test conditions

Between 7 and 12 July 2019, we captured a total of 87 rats from 3 different sites on Wake Island, including one area of natural "bush," a commensal "downtown" area where the galley and bulk of the billeting structures are located, and the SWAA where the island incinerator is located (see Figure 1). We conducted cage trials in a small wooden outbuilding previously used as storage for recreational golf equipment (hereafter "caddy shack"). Following capture, all rats were dusted with Drione® to treat for ectoparasites per standard lab test procedures (NWRC 2005). All individuals were then weighed using a digital scale, sexed, and placed singly into a plastic rodent bin (44.5 × 24 × 20.5 cm) with built-in feeding and watering functionality. Each bin was lined with approximately 120 g of corn cob bedding (Bed-o'Cobs®) and 8 g of shredded paper (Bed-r Nest®) for nesting and furnished with a small PVC tube (12 cm diameter × 45 cm length) for shelter. Daily health checks were conducted 3 times per day (~ 9 am, ~ 3 pm, ~ 9 pm) throughout the entire study period to assess health and to resupply food and water, as necessary. To minimize disturbance, consumption was recorded at the end of each phase as opposed to recording daily values. During checks, areas of excessively soiled bedding were removed and replaced with

Figure 1. Wake Atoll showing trapping locations for the "downtown" commensal area, the solid waste aggregation area (SWAA), and the "bush" rat habitat around the caddy shack.

Table 1. Room temperature log from caddy shack during the 2019 palatability and efficacy trials on Wake Island, Wake Atoll. July 7th to July 12th not recorded because the data logger could not be located.

Date	Time	Days post exposure	Temperature	Relative Humidity	Heat Index
13 July, 2019	12:40	2	30.5	77	38.3
13 July, 2019	19:50	2	28.9	78	34.0
14 July, 2019	09:00	3	28.3	84	33.6
14 July, 2019	14:45	3	28.3	79	32.7
15 July, 2019	07:35	4	27.8	78	31.3
15 July, 2019	15:40	4	31.1	73	38.8
15 July, 2019	20:20	4	28.3	78	32.5
16 July, 2019	07:40	5	28.3	80	32.9
16 July, 2019	13:30	5	30.5	71	36.6
16 July, 2019	20:40	5	28.9	78	34.0
17 July, 2019	07:55	6	28.3	79	32.7
17 July, 2019	12:50	6	31.1	72	38.5
17 July, 2019	20:00	6	28.3	78	32.5
18 July, 2019	06:00	7	27.8	79	31.5
18 July, 2019	12:30	7	31.1	73	38.8
19 July, 2019	07:45	8	28.3	76	32.2

fresh bedding ("spot cleaning"). Due to temporary inability to locate data loggers, we were only able to collect temperature and relative humidity data in the caddy shack starting 2 days after the exposure period was initiated, for a total of 7 days. Temperatures and the heat index from the final two days of the exposure period and entire post-monitoring period averaged 29.0 ± 1.3 °C (range: 27.8–31.1 °C) and 34.4 ± 2.8 °C (range: 31.3–38.8 °C). The weather and temperatures on Wake Island during the time when temperature and humidity were not recorded were like those we experienced when the data were logged (Table 1) and were unlikely to be significantly different from the range of temperatures and heat indices we recorded.

Head-to-head two-choice efficacy and palatability trial and no-choice efficacy trial

Acclimation phase

After capture, all rats were acclimatized to captive conditions for at least 2 days (maximum 4 days) before exposure to test materials. During acclimation, all rats were given ad libitum access to LabDiet® 5001 Rodent Diet maintenance feed blocks and water. Two rats died during the acclimation period and were replaced with two animals from contingency/spare animals maintained for this purpose.

Exposure phase

A 4-day exposure period was selected to emulate the critical period of bait availability for all rodents following a single aerial bait application during eradication operations (Broome et al. 2017). Fifteen adult rats from each of the 3 different capture sites (n = 45 total) were randomly assigned to the two-choice treatment group (7 or 8 of each sex per location), and 5 individuals from each location (n = 15 total) were assigned to the no-choice trial (2 or 3 of each sex per location). During the study we discovered that one of the females from the bush site in the two-choice trial was pregnant. We set an acceptable threshold of ≥ 80% mortality for the no-choice trial to assess efficacy of FINAL soft bait. Because captures in the bush and downtown areas were sparse, 10 rats captured at the SWAA were randomly selected to serve as controls for both trials. Before the exposure phase began all rats were moved into new bins with fresh bedding, cob, and water bottles.

For the two-choice treatment group, 20 g of B-25W pellets were weighed using a digital scale and scattered on the bin floor, along with a single sachet of FINAL soft bait (~ 15 g). Rats in the no-choice group were only given a single sachet of FINAL. Depleted pellets or sachets were replenished so that all rats had ad libitum access to their respective bait types throughout the exposure phase. All additional bait was weighed and recorded before being added to a cage. Because baits can change weight based on environmental conditions, 3 separate samples of each bait type were weighed and prepared in similar quantities and exposed to ambient room temperature/moisture. Changes in their weight were used to generate correction factors for consumption estimates. At the end of the exposure phase, all uneaten or spilled bait was separated from any bedding and weighed.

Observations were recorded for any rats found to exhibit symptoms of rodenticide intoxication. If at any time a rat was observed to be exhibiting signs of severe pain and distress, criteria for euthanasia as a humane endpoint would be evaluated and interventional euthanasia employed, but no animals met the criteria for interventional euthanasia. Mortalities were removed immediately and weighed using a digital scale. We did not perform autopsies on mortalities because it is unclear that visual inspection would necessarily be sufficient to determine if death occurred due to anticoagulant toxicity.

Post-exposure monitoring

Following the exposure phase, all bait was removed, and bins were cleaned and replenished with fresh bedding, cob, and water. All baits were replaced with approximately 40 grams of LabDiet 5001® Rodent Diet maintenance feed blocks during the post-exposure phase and replenished ad libitum. Surviving rats were monitored for at least 2 days (maximum 4 days) to enumerate subsequent mortalities resulting from rodenticide intoxication. Although a postexposure monitoring period of 14–21 days is generally recommended for assessing the efficacy of brodifacoum anticoagulant rodenticides (EPPO/OEPP 1999), our monitoring period was necessarily abbreviated due to logistical constraints. Although it is possible that additional mortalities may have been observed if the post-exposure monitoring period was longer, our objectives were primarily to evaluate consumption during the exposure period. Mortality estimates should therefore be interpreted as minimums. At the end of the post-monitoring phase, all surviving rats were euthanized.

Statistical analyses

All statistical analyses were performed using R 3.5.2 (R Core Team 2018) and figures were produced using the package ggplot2 (Wickham 2009). All statistical tests were two-tailed with significance levels of $p < 0.05$. The percent mortality within treatment groups and the control group was compared using a Chi-square contingency test. Differences in bait preference among treatment groups was assessed using ANOVA. All tests validating assumptions were performed. Means are reported with ± 1 standard deviation of the estimate.

Results

Control group

There were no mortalities in the control group that was not exposed to brodifacoum. Average weight change in this group was −1.2 ± 3.8 g (range: −4.1–3.1 g) for males and −1.2 ± 4.6 g (range: −8.8–4.0 g) for females.

Two-choice trial: palatability

B-25W pellets were clearly preferred, regardless of capture location, as none of the rats in the two-choice trial consumed any of the FINAL Soft Bait (Supplementary material Table S1). At the end of the four-day exposure period all FINAL sachets were still intact with no bait taken. The mean body mass of all rats at the initiation of the diet trials was 58.7 ± 9.2 g (range: 35.5–73.7 g; n = 22) for males and 53.3 ± 8.3 g (range: 35.7–73.4 g;

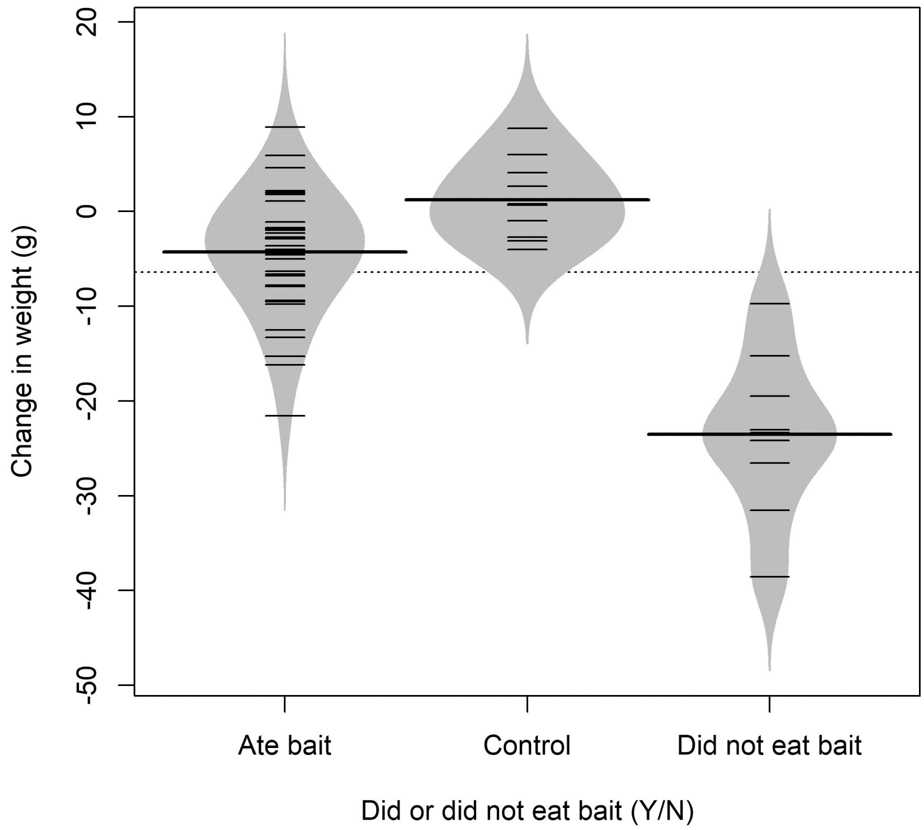

Figure 2. Distribution and mean change (longer line) in weight of rats that ate pellets of the toxic B-25W bait and rats that did not eat any bait before dying. Changes in weights of control rats from start to end of the study are provided as comparison.

n = 23) for females. There was no significant difference in body mass of male or female rats among capture locations (one-way ANOVA both $p > 0.27$) or treatment groups (one-way ANOVA both $p > 0.90$).

Two-choice trial: efficacy

The first 3 mortalities occurred 2 days after toxic bait was provided to the treatment groups. However, of these mortalities only 1 subject from the SWAA ingested bait. The other 2 individuals that died were both from the bush location and had not ingested toxicant as both the B-25W pellets and soft bait were not consumed. Unexpectedly, 8 of the 44 mortalities (18%), including 5 rats from the bush habitat surrounding the caddy shack (Figure 1) and 3 rats from the downtown location (Figure 1), did not consume any bait at all. An additional individual from the downtown area also did not consume either bait product and was euthanized at the end of the trial.

Because we could not attribute these 8 mortalities to ingestion of toxicant, we suspected they may have been due to starvation. We compared the change in weight in individuals that died but did not eat any bait with those that died but consumed some amount of the pellet formulation. Individuals that did not eat either formulation of bait lost significantly more weight ($\bar{x} = -23.5 \pm 8.5$ g) than those that consumed the pellets ($\bar{x} = -4.3 \pm 6.5$ g; two sample t-test, $t = -7.5$, d.f. = 43, $p < 0.001$; Figure 2).

There was no difference in weight loss between rats from the bush ($\bar{x}$ = −23.8 ± 11.7 g) and downtown ($\bar{x}$ = −23.1 ± 2.9 g) locations that died but did not consume either bait (two sample *t*-test, $t = -0.12$, d.f. = 7, $p = 0.9$). Given that the average initial weight for the individuals that did not eat bait was 57.2 ± 10.1 g, a change in weight of the magnitude of 23.5 g represents a 41% decrease in weight.

Because no rats in this trial consumed FINAL, we can state that the overall efficacy of B-25W was 80% (36 of 45) when including all non-eaters. Regardless of capture location, mortality was 100% for the 36 rats that did ingest B-25W pellets (Table S1), including 15 of the SWAA subjects, 10 of the bush subjects, and 11 of the downtown subjects. For the rats that ingested B-25W, the average time to mortality was 4.4 ± 1.3 days (range: 2–7 days) after the initial offering for rats from the bush and SWAA locations, and 5.2 ± 1.2 days (range: 3–7 days) for rats from the downtown location; differences among locations were not significant (one-way ANOVA: $F_{2,42} = 2.22$, $p = 0.12$). On average these 36 rats ingested 18.0 ± 5.0 g (range: 1.8–30.4 g) of the B-25W pellets.

No-choice trial: efficacy

Of the rats in the no-choice trial (FINAL Soft Bait only), 8 of 15 (53%) succumbed by the end of the abbreviated monitoring period. Like the two-choice trial, some individuals in the no-choice trial did not consume any bait ($n = 5$; 2 from the bush and 3 from downtown). Two other individuals ate some bait (range: 3.1–8.6 g) but were still alive on the last day of monitoring (Table S1). Mortality was 80% (8 of 10) for rats that consumed any amount of FINAL bait. On average, time to death for individuals that consumed bait in this trial averaged 6.1 ± 1.5 days (range: 4–8 days) following initial exposure to the bait. These individuals ingested an average total of 16.6 ± 3.2 g of bait, roughly the equivalent of a single sachet of bait.

Discussion

Palatability

When presented with a choice of toxic brodifacoum bait formulations, rats inhabiting the natural bush, downtown, and SWAA areas on Wake Island overwhelmingly preferred the hard pellet formulation (B-25W) over the soft bait formulation presented in a sachet (FINAL). None of the rats presented with a choice between the two baits consumed any of the FINAL Soft Bait. Furthermore, 5 of the 15 rats that were only presented FINAL did not consume any of this bait, despite this being the only food option available during the 4-day exposure period. Somewhat unexpectedly, 9 rats in the two-choice trail did not consume either bait; however, all rats that consumed B-25W pellets died. These results indicate that even if rats from the downtown commensal and SWAA areas on Wake Island have dietary or sensory preferences, FINAL Soft Bait does not satisfy their preferences. Based on these results, FINAL Soft Bait is a poor alternative bait to consider for application on Wake Island where aerial broadcasting of pellet formulations may be restricted due to permanent human habitation.

Given that rats are known to prefer high-fat/high-sugar food items, particularly when food is limited (Lucas and Sclafani 1989; Warwick and Schiffman 1992; Sclafani 2001), it is somewhat surprising that none of the rats in the two-choice trial consumed the FINAL bait, even though both reviews of the 2012 failed attempt to eradicate Polynesian rats from Wake Atoll identified the possibility that rats in the downtown commensal and SWAA areas could be conditioned to alternative food sources (Brown et al. 2013; Hanson et al. 2019). It is possible that the protocols regarding foods and food waste put in place in response to the failure have reduced or eliminated food "leakage" from the waste disposal chain to the point that rats have not become conditioned to alternative food sources that the FINAL bait is attempting to mimic. Another possibility is that the FINAL bait does not mimic the alternative food sources closely enough. After all, rats preferred B-25W when given a choice of alternative naturally occurring food sources (Shiels et al. 2015) and, in this study, FINAL Soft Bait. Our current knowledge indicates that rats on Wake Island may prefer the pellet formulation of B-25W over both FINAL and some naturally occurring food items, but we would need to repeat the Shiels et al. (2015) study using rats inhabiting the different habitat types to address this question more fully.

Rats may also have avoided the FINAL bait because it was an unfamiliar food. Although B-25W would also have been novel to the rats in this study, the pellet formulation of B-25W may not have represented as novel a food item, being closer to naturally occurring food items in sensory properties (e.g., hard, and dry). Food neophobia is well documented in rats (Barnett 1975; Inglis et al. 1996; Morón and Gallo 2007) and is typified by the initial avoidance of unfamiliar food, followed by gradual sampling over time and if the item does not make the rat sick it will increase consumption (Barnett 1975). To our knowledge, soft baits in sachets have never been used on Wake Island, so perhaps rats were simply not familiar with this novel food item and only the rats in the no-choice trial were given enough incentive to consume it. However, a recent study did not find support for rats demonstrating food neophobia but did document fear of eating from novel containers (Modlinska et al. 2015), a result shared by other studies (Mitchell 1976; Inglis et al. 1996). Unlike the pellets that are freely available to rats following broadcast application, the soft bait is contained in thin plastic sachets. It is possible that the novel sachet wrapper could have triggered an initial aversion to the FINAL baits in our trial. This does not seem likely though, particularly for the rats inhabiting the SWAA that are presumably more frequently exposed to food resources in novel packaging. Norway rats (*R. norvegicus* Berkenhout, 1769) inhabiting landfills and other highly variable environments are known not to exhibit neophobia, presumably because their environment was constantly changing, and they regularly encounter novel food items (Barnett 2004; Modlinska and Stryek 2016). The SWAA shares these characteristics, so presumably rats in this area should be conditioned to be more indiscriminate in their preferences. This study is the only one we know of testing FINAL or any other soft rodenticide bait sachet in a field study, so it is difficult to know if this is a global issue or unique to the rat population on Wake Atoll. Without further experimentation, it will be difficult to know the ultimate mechanism behind this result, but for the purposes of a future rat eradication attempt on Wake Atoll, the lack of palatability with rats from Wake Island makes FINAL an unlikely alternative to the pellet formulation.

We also documented 9 individuals that did not consume the pellet bait, even when no other palatable alternatives were available. A previous palatability study on Wake Island also documented individuals avoiding the pellet bait (Shiels et al. 2015). While this could be the result of any of the reasons covered in the neophobia discussion (see above), it is interesting to note that these individuals came from the downtown commensal area and the bush habitat around the caddy shack. Both areas of the island experience intermittent to regular rodent control using another rodenticide, CONTRAC® All-Weather Blox, that uses the anti-coagulant bromadiolone (Wake Island pest control personnel, *pers. comm.*). Some researchers have suggested that food neophobia could result from attempts to control or eradicate rats (Barnett 1975; Taylor and Thomas 1989; Inglis et al. 1996), which may explain why rats from these habitats avoided the pellets but not the rats from the SWAA. This is a cause for concern moving forward; while most of the individuals that did not consume bait subsequently died, the magnitude of their change in weight, on average > 20 g or approximately 40% of their overall body weight, indicates that they likely starved even though they had access to bait during the 4 days of exposure. Hunger usually acts as an impetus for rats to overcome their neophobia and consume food (Barnett 1975), but if these individuals were in a natural setting, they would have had access to alternative food sources and presumably would have ignored the pellet bait and survived. This raises the possibility that rats in these subpopulations may be pre-conditioned to avoid pellet bait, highlighting the importance of suspending rodenticide use well in advance of a proposed eradication and offering alternative bait options throughout the course of the bait efforts (Broome et al. 2017).

Another possible explanation for why some individuals in each of the treatment groups may not have eaten either bait product is the temperature in the non-climate-controlled caddy shack, where the trails were performed. Our study was conducted during the warmest and most humid period of the year (USAF 2017), and even with fans running continuously during the study we recorded afternoon temperatures above

30 °C (Table 1). Combined with the relative humidity, there were times during almost every day of the study where the heat index for the room could be over 40 °C (> 100 °F). Hamilton (1963) found that rats exposed to temperatures above 35 °C decreased their food intake to as low as 2 g compared to earlier intake of 20 g at 24 °C, and that even mild heat stress (32 °C) over time continued to lower food intake. Rats in our study appeared to be sluggish during the mid-day, so perhaps heat stress may have caused some individuals to lose their appetite. However, most subjects in our study consumed some combination of lab chow and/or baits and there were no mortalities in the control group during the study. If the temperature did impact food intake it clearly did not affect all the animals in the same manner.

Efficacy

As previously discussed, some of the mortalities in the no-choice trial to assess the efficacy of FINAL on the Wake Island rats were likely due to starvation versus toxicosis. Given that these individuals did not consume any bait, these mortalities cannot be attributed to the toxic effects of the bait and were not used to assess the efficacy of FINAL. When these individuals were removed from the analysis, 80% of the rats that ingested bait perished before the end of the abbreviated monitoring period, indicating that this bait is effective when consumed. The two individuals who survived the trial and subsequent 4-day monitoring period consumed the smallest amount of bait (< 9 g). Unfortunately, we only recorded when bait was made available to the rats but did not record when rats initiated consuming the bait. Like Shiels et al. (2015) we noted that some rats delayed eating bait, so the average of 6 days for mortality to occur in this trial includes time when rats may not have ingested any rodenticide. A previous efficacy study of brodifacoum on *R. exulans* found mortality occurred within 4–9 days following exposure (Pitt et al. 2011). If either of the two individuals that consumed bait, but did not die, delayed eating until the last day bait was available, it is possible, given the short 4-day post-exposure monitoring period that these individuals may have ingested a lethal dose but did not have sufficient time to succumb to toxicosis during the brief post-exposure monitoring phase. One of these individuals did display symptoms often displayed by other rats prior to mortality from ingesting a lethal dose (e.g., hunched posture, unkempt coat, and diarrhea) two days prior to being euthanized at the end of the study.

Like the no-choice trial, once individuals from the two-choice trial that did not consume either bait were removed from our efficacy analysis, B-25W was highly effective (100% mortality). Because none of these animals consumed any of the FINAL bait, this trial effectively operated as a no-choice efficacy trial for B-25W, which is the most direct test of efficacy.

While we cannot say for sure what caused the mortality of the individuals in both treatments that did not consume any bait, it seems highly likely that it resulted from starvation, potentially exacerbated by heat stress. The average weight loss in these individuals was 41% of their body weight, versus only 7% in the individuals that consumed bait and presumably died from toxicosis, while the control group gained weight. We would expect for rats that ingested a lethal dose of bait to lose some weight, as most individuals do go through some period of lethargy and cease eating before dying. However, this seldom takes more than several days, and weight loss is rarely as dramatic as what we observed in the animals that we know did not consume any bait during the study.

Conclusions

The potential for dietary and sensory preferences of target rat populations was identified as a possible factor contributing to the failure to eradicate Polynesian rats from Wake Atoll in 2012. Identifying alternative bait formulations that address these preferences and are suitable for application techniques other than aerial sowing may provide better exposure of rats to bait for a future eradication attempt. Efficacy trials indicated that FINAL Soft Bait by Bell Laboratories had an 80% mortality rate for rats that ingested the bait. However, when offered B-25W pellets as an alternative, rats from different areas on Wake Island did not consume any FINAL bait. These results indicate that rats clearly preferred the pellet formulation, and do not support the hypothesis that localized sub-populations might have unique food and/or sensory diet preferences for soft baits. As such, FINAL is not a recommended alternative to the B-25W pellet formulation on Wake Island. While these results were unequivocal and are useful for planning of future eradication attempts on Wake Atoll, we caution about extrapolating them to include other islands and rat populations, as local environmental conditions and population preferences likely differ from those on Wake Island.

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Ethics and Permits

All animal use was conducted according to protocol QA-3075, approved by the NWRC Institutional Animal Care and Use Committee.

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Supplementary material

The following supplementary material is available for this article:

Table S1. Summary of rat capture data from Wake Island, Wake Atoll.

This material is available as part of online article from:
http://www.reabic.net/journals/mbi/2021/Supplements/MBI_2021_Kappes_Siers_SupplementaryTable.xlsx