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# Large-scale Aerial Baiting to Suppress Invasive Rats in Hawaii: Efficacy of Diphacinone and Associated Risks

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**ABSTRACT:** Invasive rats are among the most damaging animals to native species on many island ecosystems including those in Hawaii. On Oahu Island, U.S. Army Garrison Natural Resources Program currently manages invasive rat populations to protect natural resources by using grids of A24 automated traps, and previously snap-trap grids and rodenticide bait stations. Despite these control efforts generally suppressing rats, some lands with natural resources that are at risk to rat predation are not easily accessible for implementing these traditional rat control methods. In a 430-ha mesic forest on Oahu where ungulates are excluded and site access is limited due to military training and presence of live ordnance, we tested the efficacy of aerial application of anticoagulant rodenticide bait pellets (Diphacinone-50 Conservation), applied in two applications at a rate of 12.82 kg/ha per application. We measured the effectiveness of the rodenticide bait application by deploying tracking tunnels (inked and baited cards to identify rat presence) before, during, and after applications within treated and nearby untreated areas. Due to restricted access, we failed to estimate nest success of an endangered bird; yet previous research showed rat control increases this bird's population. We also measured diphacinone residues in stream water at the treatment site to determine this method's risk level to the aquatic ecosystem. The aerial application resulted in immediate and sustained reduction in the rat population, as evidenced by rat activity decreasing from ~44% to 3.8% during the first three months after bait application and maintained <20% rat activity for 10 months. Trail cameras and recovered rat carcasses also highlighted effectiveness. One of 34 stream samples analyzed had detectable diphacinone residues and this single sample was taken one week after application and it had very low levels of diphacinone (below levels quantifiable). Aerial application of diphacinone appears to be an efficient and effective rat suppression technique for natural resource protection in complex landscapes.

**KEY WORDS:** aerial-broadcast baiting, diphacinone rodenticide, endangered species, helicopter application, island invasive pest species, native biodiversity, *Rattus*, rodent management, tracking tunnels

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## INTRODUCTION

Invasive rodents [i.e., rats (*Rattus* spp.) and mice (*Mus musculus*)] occupy most of the world's land masses, and they are particularly damaging to natural resources (i.e., native species) on islands (Towns 2009, Witmer and Shiels 2018). Their introductions to new islands or ecosystems generally occur unintentionally and via human transport; however, new rodent introductions to islands can also result from large storms such as hurricanes (Shiels et al. 2020). Eradications of rats and mice are generally not possible on islands that are exceptionally large (e.g., the main Hawaiian Islands) or heavily comprised of humans and private lands; therefore, invasive rodent control or suppression through repeated trapping and/or toxicant use within segments of islands are the most common forms of protecting native species from the negative impacts of rats and mice on islands (Duron et al. 2017).

Invasive rats, primarily *R. rattus* (black rat) and to a lesser degree *R. exulans* (Pacific rat), have been documented as important predators of native and endangered species in Hawaii, including birds, arthropods, snails, and plants (Meyer and Shiels 2009, Shiels et al. 2013, Shiels et al. 2014, Banko et al. 2019). In 2000, U.S. Fish and Wildlife Service (USFWS) granted the Oahu ʻelepaio (*Chasiempis ibidis*), a native forest bird endemic to Oahu, endangered species status under the federal Endangered Species Act of 1973. Due to the elevated rates of rat predation on Oʻahu ʻelepaio eggs and chicks (VanderWerf 2001, VanderWerf and Smith 2002), as well as on endemic snails and plants (Meyer and Shiels 2009, Shiels and Drake 2011, Pender et al. 2013), the Army's Natural Resources Program on Oahu (ANRP) has been engaged in rodent control since 2001 using various techniques including snap traps, automatic traps, and rodenticide bait stations. The Lihue Management Unit (LMU) is one of these areas where ANRP controls rats for the benefit of ʻelepaio nesting success as well as to promote additional native plants and animals that are at risk to rat predation.

In many Hawaiian forests including those on Oahu, isolated populations of rare plants, rare snails *(Achatinella mustelina)*, and the Oahu ʻelepaio have been supported with a system of small grids of traps and/or bait stations in the attempt to control rat predation on these rich natural resources. Limited access and the labor-intensive nature of servicing these traps and bait stations means that, in general, they may only be re-baited every 2-6 weeks. However, the use of Goodnature A24 self-resetting rat traps has greatly extended the trap-servicing requirements to maintain suppressed rat populations (Shiels et al. 2019a). Despite these improvements, some locations still require additional rat suppression to successfully protect native and endangered species. Additionally, in areas with substantial amounts of unexploded ordnance (UXO), rat trapping grids may not be realistic or efficient due to the limited land access and repeated needs to service the traps. Thus, aerial rodenticide treatment (via helicopter) is of interest over a large portion of particular sites on Oahu, including the LMU. This would be a one-time (2 application) of Diphacinone-50 rodenticide bait just prior to the nesting season of Oahu ‘elepaio to determine the efficacy of such a method in the LMU and other areas that need protection from rat predation.

The overall objectives were: 1) monitor rat activity and fate before, during, and after the rodenticide application, 2) document the non-target effects through trail cameras and carcass searches, and 3) sample the water from the stream running through the study site and test it for diphacinone residues before, during, and after the rodenticide application. The efficacy of the bait application benefiting the Oahu ‘elepaio nesting success was monitored by ANRP staff that had been monitoring these bird populations in the LMU for the previous 10+ years. Based on hand-broadcast studies in 2015-2016 that used this same bait applied in smaller portions of mesic forests in the Waianae Mountains (Shiels 2017, Shiels et al. 2019a), we hypothesized that rats would be the main animal consuming and suffering from the bait application, and reductions in invasive rats would occur using the planned Diphacinone-50 application.

**Figure 1. The Lihue Management Unit (LMU), which includes the forested mountains (Waianae Mountains, O‘ahu) in the background. The foreground, and the midground on the right side of the photograph, represent live-firing ranges that are used by the U.S. Military. The LMU is home to many threatened and endangered species, including the O‘ahu ‘elepaio (*Chasiempis ibidis*).**

## METHODS

### Study Site

The LMU is located at 600-1000 m elevation in the Waianae mountain range, within the western portion of Schofield Military Base, on Oahu Island, Hawaii. The LMU is a large area (714 ha) with steep terrain (Figure 1), yet the greatest restrictions in managing the natural resources in the LMU are that 1) substantial amounts of UXO are present, and 2) it is located on an active Army training range that is only accessible to natural resource managers 4 to 5 days each month. A 430-ha area within the LMU was used for this study.

The LMU is covered in mesic forest and it is home to many rare taxa, including Oahu ‘elepaio, plants, and snails. Non-native rodents are ubiquitous at LMU, including black rats, Pacific rats, and house mice; black rats are numerically dominant in the Waianae Mountains (Shiels and Drake 2011, Shiels et al. 2013).

### Aerial Application of Rodenticide

The aerial application method for reducing the rat population just prior to the nesting season of Oahu ‘elepaio is through a two-application (“one-time”) aerial broadcasting of ‘Diphacinone-50: Conservation’ according to label (Diphacinone 50: Conservation, EPA Reg. No.: 56228-35). The entire project has been through the NEPA process, headed by U.S. Army Garrison NEPA Coordinator (EA evaluated, public comment period, EA signed by Commander; see Supplemental EA Finding of No Significant Impact 2017).

Aerial broadcast application of Diphacinone-50 involved a contract pilot that was certified for aerial application of restricted use pesticides in Hawaii. To apply the bait, a helicopter fixed with a specialized suspended bucket flew along predetermined Global Positioning System (GPS)-plotted transects within the application area (Dunlevy et al. 2000). The 430-hectare (ha) treatment area was contained within a fenced enclosure located in the 714 ha LMU. Diphacinone-50, Pelleted Rodenticide Bait, containing the anticoagulant rodenticide diphacinone (0.005% active ingredient) has been approved for aerial distribution by the U.S. Environmental Protection Agency (EPA) and the Hawai’i Department of Agriculture (HDOA) (EPA Reg. No.: 56228-35, State of Hawaii Lic. No. 8600.1). The first application spanned two days, November 28-29, 2017, and after five days (i.e., on December 3, 2017) a second application at the same rate followed. The bait rate for each application was targeted at 11.1-13.8 kg/ha, in accordance with the approved Diphacinone-50 label. Shiels and all ANRP applicators were certified with Hawaii restricted pesticide category 2 (Forest Pest Control) at the time of the operation.

### Rat Population and Environmental Monitoring

Rodent populations were monitored before, during, and after diphacinone treatment in the 430-ha treatment area and in an adjacent untreated area (reference site, known as Coffee Gulch). We used 150 tracking tunnels (120 in treatment area, 30 in reference area), which are baited ink cards placed in tunnels so that footprints of animal visitors can be identified, to determine presence of invasive rodent species at each location. Tracking tunnels (50-cm-long black corrugated plastic tunnels with 10 × 10-cm openings at both ends), were baited with peanut butter, and were set for one night per sampling period. Quarterly assessments of rodents at both the reference site (Coffee Gulch) and treatment site (LMU) occurred during the year leading up to bait application. Rodent monitoring using tracking tunnels also occurred: one day prior to first application, 1 day prior to second application, three weeks after $1^{st}$ application, and monthly thereafter for at least six months. An acceptable level of rat activity, which promotes stable or increasing native/endangered Oahu ʻelepaio populations has not been clearly identified, but New Zealand studies have shown that rat activity levels of 10% are low enough to maintain certain rare bird populations (Innes et al. 1999). A 10% activity level, and at times even a 20% activity level (which was shown beneficial in the Waianae Mountains for conserving endangered plants; Pender et al. 2013, Shiels et al. 2019a), has not always been achievable using large-scale rat trapping in Hawaii, indicating a further need for testing rodenticides in this manner.

In addition to estimating the changes in the rodent populations following rodenticide treatment, the effectiveness of the treatment was also monitored by conducting carcass searches and using trail cameras to determine the animals consuming and removing the bait pellets. Carcass searches occurred before, during, and after bait application, and involved walking trails at LMU and the reference site while visually scanning the ground for any rodent and other vertebrate carcass within approximately a 2-m swath on either side of the trail. During the five days following the first bait application, 20 Reconyx (Hyperfire model PC900) motion-sensing cameras were positioned 1 m from bait pellets (two pellets per station) to document the types of animals visiting the baits. Cameras were programmed to obtain bursts of three still-photos per triggering event. The bait application rate from aerial broadcast was measured on the ground, and, like the condition of the bait on subsequent days after both applications, are reported in Shiels et al. (2019b).

### Stream Water Monitoring

Water, collected from the stream running through the study site, was collected before, during, and after bait application and analyzed for concentrations of the diphacinone compound. Sampling schedule followed that of the tracking tunnels mentioned above. Stream water samples were analyzed by using an Agilent 1100 liquid chromatograph (HPLC) at USDA NWRC. We identified the Detection limits (DL), which is the concentration of diphacinone required to generate a detectable signal using this method, and Quantitation limits (QL), which is the concentration of diphacinone required to be able to reliably quantify the amount of diphacinone in the sample. Thus, if a sample had concentrations above the detection limit, but below the quantification limit, then care should be taken when evaluating the results because the variability will be significantly greater than the quality control samples. The DL and QL levels used in this study for stream water were DL = 0.0035 ng/g and QL = 0.012 ng/g, respectively.

## RESULTS

### Bait Applied from Helicopter, Bait Cost, and Remaining Bait

To the 430 ha LMU, we applied bait at a rate of 12.13 kg/ha for the first application and 13.55 kg/ha on the second application; both bait applications were under the maximum of 13.8 kg/ha stated on the EPA label for Diphacinone-50. Our bait applications resulted in approximately 1 bait pellet per $m^2$ (see below). Because each pellet was approximately 1.1 g, there were roughly 260,000 pellets applied per application. In total, 26,750 lbs (12,133 kg) of Diphacinone-50 was ordered from Hacco (Neogen Corporation), which costed $31,104.90 (535 units at $58.14 each), and $11,000 in shipping from Wisconsin.

### Rat Population and Environmental Monitoring

Rat tracking was 44.4% at the treatment site prior to bait application, and 3.8% during the three months after bait application. Rat tracking at the reference site was 42.8% prior to bait application, and 43.3% after bait application (Figure 2). At LMU, but not the reference site, we had land access to conduct monitoring during 1 May 2018-22 Oct 2018 (4 periods), and the rat tracking during that time averaged 8.5% (Figure 2). The 19 Nov 2018 rat tracking was back up near pre-treatment levels, at 42.4%. At the reference site when we finally got access again (18 Dec 2018-24 April 2019), rat tracking was 48.9%, whereas during that same period at the treatment site the rat tracking was 41 percent.

For the limited time that we had access, all trail camera photos where bait pellets were removed were by rats or mice, and there was one incidence when a game bird removed a pellet. Several other photographs of non-target species were captured after the bait application, but there were no further occasions where these animals (e.g., mongoose, birds) touched or removed the bait. Similarly, the only carcasses found in the LMU were rodents.

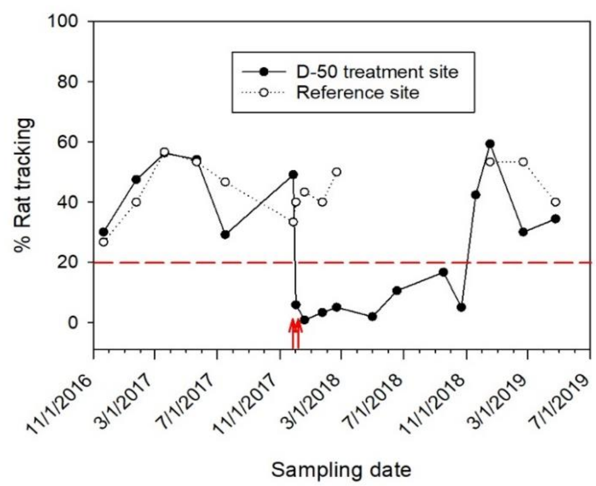


**Figure 2. Tracking tunnel results, which indicates rodent activity and population status, for invasive rats (*Rattus spp.*) at the treatment site (LMU) where Diphacinone-50 bait was applied by aerial broadcast, and the reference site where no rat control occurred. The arrows represent the two dates of bait application (28-29 November 2017 and 3 December 2017). The horizontal dashed line at 20% presence is the estimated level of suppression needed to protect natural resources from rat damage. There was no access to the reference site allowed from the 1 May 2018 to the 22 Oct 2018 sampling periods.**

### Stream Water Monitoring

One of 34 stream samples analyzed had detectable diphacinone residues [0.0051 ppb (ng/ml)] and this single sample was taken one week after the first bait application and it had very low levels of diphacinone (below levels quantifiable). This single sample originated from within the LMU, and there was some bait noted in the stream near the sampling location just two days prior to this sampling (thus, two days following the second application).

## DISCUSSION

The one-time (two application) aerial broadcast of Diphacinone-50: Conservation, at current label rate, was effective at reducing two rat species in the LMU mesic forest in Hawaii. Evidence of rat reduction from this bait application included 1) rat activity decreasing from ~44% to 3.8% during the first three months after bait application, and maintained at <20% rat activity for 10 months, whereas the reference site had rat tracking consistently at 35-60% during the year-long study, 2) only rodent carcasses were found in the treatment area following bait application while no rodent carcasses were found at the reference site during the same period that the LMU was monitored, and 3) documentation via monitoring cameras of revealed rats removing bait pellets.

To our knowledge, there have been no aerial applications of diphacinone for rodent suppression in Hawaii. However, there have been three attempted rat eradications from offshore islands in Hawaii using aerial (helicopter) broadcast of diphacinone bait. One of those aerial applications was a successful rat eradication (Mokapu Island), whereas one was unsuccessful (Lehua Island first attempt in 2006) and one is yet to be determined (Lehua Island second attempt in 2017; Shiels et al. 2020). Just four hand-broadcast applications of a similar bait product as used at the LMU's aerial broadcast have been performed in Hawaii, and these are reported in Dunlevy et al. (2000), Spurr et al. (2013), Shiels (2017), and Shiels et al. (2019a). Dunlevy et al. (2000) investigated the optimal bait application rate on Hawaii Island using inert bait pellets that contained a biomarker instead of the anticoagulant compound diphacinone, and the pellets were 6 g each instead of 1.1 g like those in our study. Spurr et al. (2013) conducted a field trial at Hawaii Volcano National Park (Hawaii Island), hand-broadcasting pelleted (6 g each) Ramik Green, which is the same formulation as Diphacinone-50, for purposes of registering the product with the EPA for hand-broadcast for rat control. Diphacinone-50 was hand-broadcasted in a 36-ha area (Kahanahaiki) in Shiels (2017), and a 5-ha area in Shiels et al. (2019a); both studies were conducted in mesic forest on Oahu like those of the LMU.

Just one of 34 stream samples had detectable levels of diphacinone, and its concentration was so low that it was not detected by analytical machinery during one of two attempts; and the 0.0051 ppb (ng/ml) was below the minimal quantitation level. Not surprisingly, this single sample was taken from a location where bait had been observed in the water on the day of application. Given that diphacinone has a fairly low solubility in water, and there was just one sample with extremely low concentration of diphacinone found despite sampling several parts of the stream during multiple occasions since bait application, the prescribed treatment of diphacinone (as performed at LMU) appears to have little impact to stream reaches within a large bait application site.

Although general (non-species-specific) anticoagulants like diphacinone have inherent risks to non-target species and the ecosystem, we found little evidence that a one-time (two application) broadcast of diphacinone over a large forested area of 430 ha had strong negative impacts on non-target species and the freshwater stream that drains the treatment site. Therefore, aerial application of diphacinone appears to be an efficient and effective rat suppression technique for natural resource protection in complex landscapes. Continued efforts to link rat removal with native species responses in continuously rat-suppressed habitats will serve as a model for local, regional, and international interest groups (public, land managers, conservation biologists, funding agencies), and will hopefully apply to future island habitats that currently suffer from establishment of non-native rodents.

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