

## Research Article

## Development and evaluation of prototype toxicant-delivery bait stations for the control of the small Indian mongoose

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### Abstract

We conducted research to develop a safe and effective toxic bait to control the small Indian mongoose (*Urva auropunctata*), an invasive vertebrate predator impacting the survival of native species in Hawai'i and in other parts of the world. A preserved fish-based bait product was found to be highly palatable to mongooses in cage trials and subsequent formulations with diphacinone (0.005%) showed promise as an efficacious toxic bait for mongooses. This product is intended for future use to control mongooses in conservation and urban areas, and as a biosecurity tool at ports of entry to address accidental introductions into mongoose-free areas. Anticipated delivery of this toxic bait is in tamper-proof bait stations. We designed three prototype bait stations constructed with polyvinyl chloride (PVC) drainage pipes and evaluated their performance in enclosure trials with wild-caught mongooses and in field trials with free-ranging mongooses. A commercially available tamper-resistant rodenticide bait station was also used for comparison to the prototypes in our trials. The goal was to develop a bait station that is readily used by mongooses, allows for bait consumption in place, prevents removal of bait, and restricts access to nontarget animals. We used a non-toxic formulation of the fish-based bait product and monitored bait station use, visitation rates, bait uptake, and spillage. All four bait station types were used by mongooses in the laboratory, and one PVC bait station design and the commercial bait station exhibited multiple mongoose visitations with minimal bait spillage in the field. We did not record any significant nontarget species interactions with the bait during the field trials. The PVC bait station design and commercial bait station are approved methods of bait delivery in the subsequent field efficacy trials under an Experimental Use Permit for the upcoming registration of “Fish-based Bait for Mongooses” with the Environmental Protection Agency.

**Key words:** bait delivery, invasive species, pest control, *Urva auropunctata*, Hawai'i, vertebrate pesticide development

### Introduction

Invasive species are considered one of the greatest threats to biodiversity in island ecosystems, primarily facilitated by intentional introductions by humans for biocontrol or economic resources (Nellis and Everard 1983; Bailey 1993; Alterio et al. 1997; Abernethy et al. 2016). Native wildlife in the Hawaiian Islands evolved in the absence of mammalian predators, and

their subsequent introductions have led to many extinctions and declines of native species (Nellis and Everard 1983; Yamada and Sugimura 2004; Hays and Conant 2007). The small Indian mongoose (*Urva auropunctata* [syn. *Herpestes auropunctatus*], mongoose) was widely introduced to Hawai'i and other areas globally including Japan (Amami Oshima and Okinawa) for biocontrol of rodent pests and poisonous snakes, which failed. They have since become well established on the islands to which they were introduced (Berentsen et al. 2018). Mongooses are a primary predator of native wildlife, especially threatened and endangered waterbirds (Underwood et al. 2013) and seabirds in Hawai'i, and threaten the continued survival of the federally listed Hawaiian Stilt (a'eo, *Himantopus mexicanus knudseni*), Hawaiian Gallinule ('alae'ula, *Gallinula galeata sandvicensis*), Hawaiian Coot ('alae ke'oke'o, *Fulica alai*), Hawaiian Goose (nēnē, *Branta sandvicensis*), Hawaiian Duck (koloa maoli, *Anas wyvilliana*), Hawaiian Petrel ('u'au, *Pterodroma sandwichensis*), Band-rumped Storm Petrel ('akē'akē, *Oceanodroma castro*) and Newell's Shearwater ('a'o, *Puffinus auricularis newelli*).

The threat of accidental introductions to other mongoose-free islands in the Hawaiian islands (e.g., Kaua'i, Lāna'i) highlights the need for a comprehensive array of control techniques to quickly respond to reported sightings or incipient mongoose populations (Pitt et al. 2015; Phillips and Lucey 2016; Berentsen et al. 2018). Multiple incidents of mongoose captures on Kaua'i near port of entry locations have been recorded in the past decade (Antaky et al. 2023). Mongooses also present a health risk to humans as hosts of leptospirosis in Hawai'i (Wong et al. 2012) and the Caribbean (Everard et al. 1976), and as a rabies reservoir on several islands in the Caribbean (Seetahal et al. 2018).

Various strategies have been used to reduce or remove mongoose populations in Hawai'i and elsewhere, including trapping and toxic baits. Trapping has been useful in reducing mongoose populations and predation in and around targeted sensitive native areas, for example in wetlands with ground-nesting waterfowl (Roerk et al. 2022). Trapping, however, is labor-intensive, expensive, and only removes mongooses from limited areas (Barun et al. 2011; Sugihara et al. 2018; Berentsen et al. 2018). Toxic baits can provide a more effective, widespread and longer-lasting approach to eradicate vertebrate pests (Howald et al. 2007; Young et al. 2012; Antaky et al. 2023). Currently, there are no mongoose-only targeted toxic baits for use in the United States. In Hawai'i, a 0.005% diphacinone wax bait block product (SLN No. HI-980005) is co-labelled for use on rodents and mongooses. However, the bait block, which was originally designed to control rodents, showed relatively poor efficacy with wild-caught mongooses in a laboratory study (Sugihara et al. 2018). There was a previously registered fresh ground beef diphacinone bait (0.00025% diphacinone, SLN Reg. No. HI-910004; Keith et al. 1989) targeted for mongooses which proved to be efficacious but required pre-mixing and frequent bait replacement in the field due to rapid spoilage. This SLN registration was eventually cancelled in 1996 (Sugihara et al. 2018).

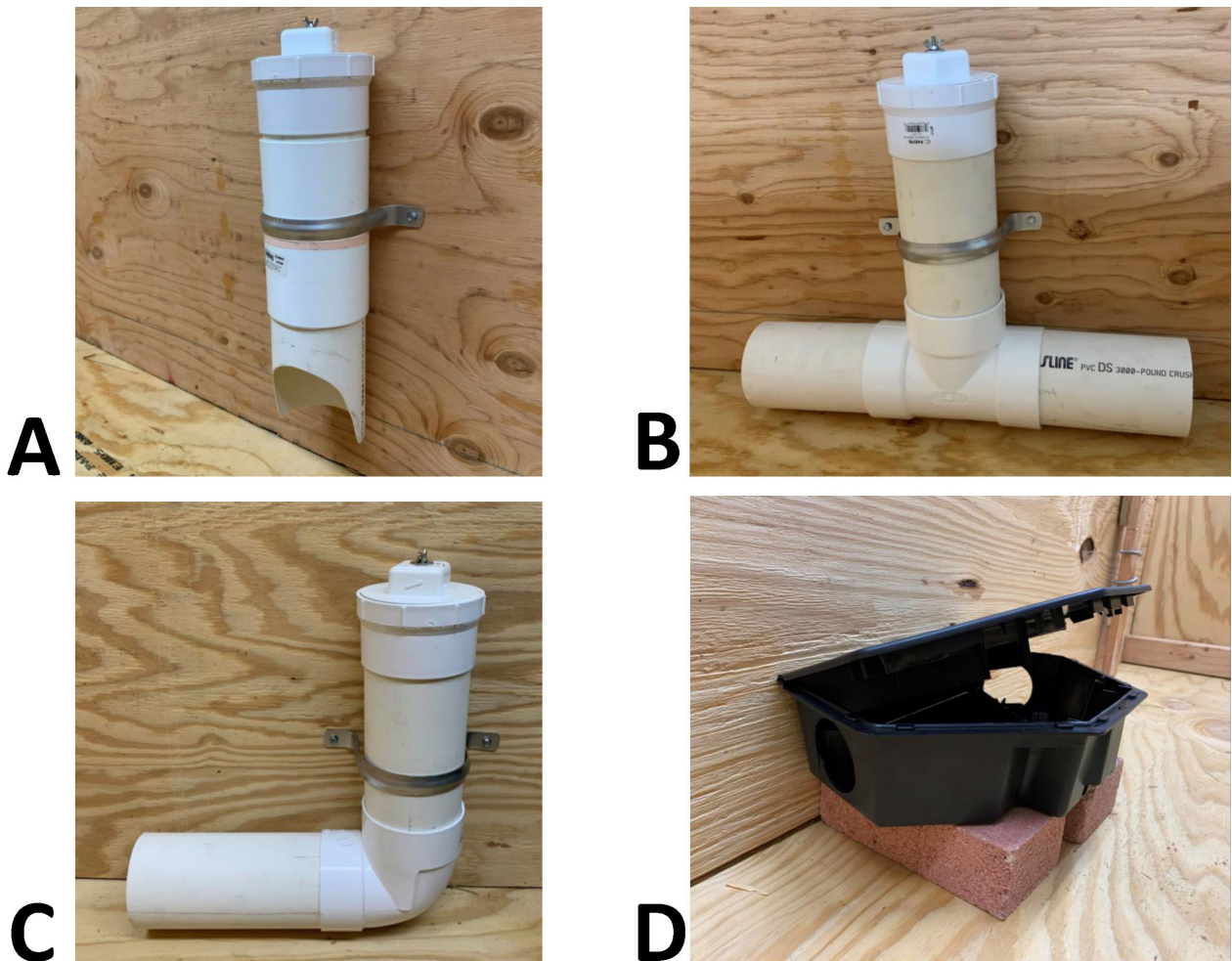
The U.S. Department of Agriculture, Animal and Plant Health Inspection Service, Wildlife Services, National Wildlife Research Center (hereafter, NWRC) has recently developed a highly efficacious mongoose control product that will soon be field tested under an Experimental Use Permit (EUP) approved by the U.S. Environmental Protection Agency (EPA) (Antaky et al. 2023). This product (hereafter, called Fish-based Bait for Mongooses) contains 0.005% of the active ingredient diphacinone within a fish-based bait matrix (Animal Control Technologies Australia (ACTA), Establishment No.: 091731-AUS-001) encased in a sausage-type biodegradable skin. This bait matrix was highly palatable in non-toxic laboratory trials of wild-caught mongoose (Siers et al. 2020). A feasibility assessment comparing the registration potential of a new toxic bait for mongooses identified diphacinone as the most promising toxicant of choice due to its relatively low cost to register and low nontarget risk (Ruell et al. 2019). The initial EPA registration for this mongoose toxic bait will be limited to use only in bait stations in terrestrial non-crop areas, which will help prevent nontarget take of the toxic bait. This type of registration, diphacinone bait product for vertebrate pests, is often delivered in tamper-resistant bait stations when used outdoors and above ground in areas accessible to the public, pets, livestock, and other nontarget species. Bait take by nontarget species can jeopardize the health of native species, and rapid consumption of bait by rats and other consumers would make the bait unavailable for mongooses that forage over large areas, especially within low density populations. To our knowledge, there are no existing commercially available bait station designs optimized for mongooses, although Keith et al. (1989) developed an inverted “T”-shaped polyvinyl chloride (PVC) bait station used to evaluate fresh hamburger diphacinone baits in field trials.

The objective of our study was to design an inexpensive, easy to deploy, effective bait station that targets mongooses, reduces consumption by rats (the primary bait co-consumer), and excludes other nontarget vertebrate species, including native birds. Candidate bait station prototypes designed for mongooses and a commercially available, tamper resistant, rat-sized bait station were evaluated in laboratory enclosure and field studies using a nontoxic formulation of the fish-based bait. The most practical bait station design(s) that performs well in these trials by demonstrating high mongoose accessibility, low bait spillage, and low nontarget bait interaction and consumption, will be selected for the upcoming EUP field trials progressing towards the goal of a registered toxic bait and delivery system for the control of mongooses.

## **Materials and methods**

### *Bait station designs*

The novel bait station prototypes designed for mongooses that were tested in this study included a raised PVC tube design (prototype A), an inverted “T”-shaped PVC design with two entries (prototype B), and an “L”-shaped



**Figure 1.** Mongoose bait station types evaluated in the laboratory study: a raised PVC tube design (prototype A), an inverted “T” PVC tube design with two entry points (prototype B), an “L” PVC tube design with a single single-entry point (prototype C), and a raised Protecta® LP commercial rodenticide bait station (type D). Photographs by NWRC Hawaii Field Station.

PVC tube design with a single-entry point (prototype C) (Figure 1). The Protecta® LP (Bell Laboratories, Madison, WI), which is a commercially available, tamper-resistant, low profile, rat-sized, rodenticide bait station (type D), was also evaluated.

#### *Test bait*

The non-toxic fish-based bait matrix was manufactured by ACTA in Somerton, Victoria, Australia based on the bait matrix for ACTA’s Foxshield® bait, which contains sodium fluoroacetate (commonly known as 1080) and is registered to control foxes in Australia. The fish-based bait matrix is compressed into cylindrical sections (~ 35 grams) and encased using a biodegradable cellulose skin (Supplementary material Figure S1). This bait matrix was previously shown to be very palatable for mongooses in controlled non-toxic, two-choice laboratory palatability trials (Siers et al. 2020).

#### *Laboratory enclosure trials*

Wild mongooses were captured from a forested habitat in Hilo, Hawai‘i, island of Hawai‘i using Tomahawk cage traps (Tomahawk Live Trap,

Hazelhurst, WI) following approved procedures (NWRC SOP AC 005.01 – Capture, handling and care of mongooses at the Hawai'i Field Station). Upon arrival at the NWRC Hawai'i Field Station in Hilo, mongooses were dusted for ectoparasites with permethrin dust. Each mongoose was then assigned an individual ID number, sexed, and weighed. During the pre-test period, animals were individually housed in 42 cm (tall) × 61 cm (wide) × 64 cm (deep) grated-bottom, stainless steel, modified rabbit-type cages (3,904 cm<sup>2</sup> floor area) mounted on metal cage racks.

Mongooses were acclimated to laboratory conditions for a minimum of three consecutive days pre-test with *ad libitum* access to bottled water in a glass bottle fitted with a stainless-steel ball-type sipper tube. Animals were offered the maintenance diet of dry cat food (approximately 70 grams per day), supplemented by 50 grams of previously frozen raw chicken parts once every 3–4 days. During the last day of the pre-test period, the pre-test diet was removed to allow mongooses to undergo a 24-hour fast. Fasting allowed the mongooses to seek out the bait quicker, which increased the likelihood of bait station visitations within the first 12 hours of the test period. Each animal was checked for visible symptoms of health or lethargy at least twice daily (morning and afternoon) during the acclimation period.

After ≥ 3 days of pre-test acclimation, mongooses were transferred to one of four artificial test enclosures created in a separate animal room (Figure S2). Enclosure walls were constructed of 5/8–3/4 in (1.6–1.9 cm) thick untreated plywood sheathing coated with a moisture sealer (4 ft × 8 ft or 120 cm × 240 cm) secured together with metal wood screws and hinges for easy disassembly and transport. An entry/exit door was constructed on one end of the enclosure to facilitate transfer of mongooses and ease of daily maintenance of the enclosure. Each enclosure included a floor space of 32 ft<sup>2</sup> (~ 3 m<sup>2</sup>) with a 3 ft (~ 1 m) high wall. The floor of each enclosure was constructed with plywood and lined with dried grasses. Metal mesh (chicken wire) was secured to the top of the enclosure to prevent escapes. A standard PVC drainage pipe 12 in (~ 30.5 cm) length and with a 6 in opening (~ 15 cm inside diameter) was provided in each enclosure to provide refuge. A water bottle was attached to the wall of each enclosure and water was available *ad libitum*. Each enclosure was assigned a unique number.

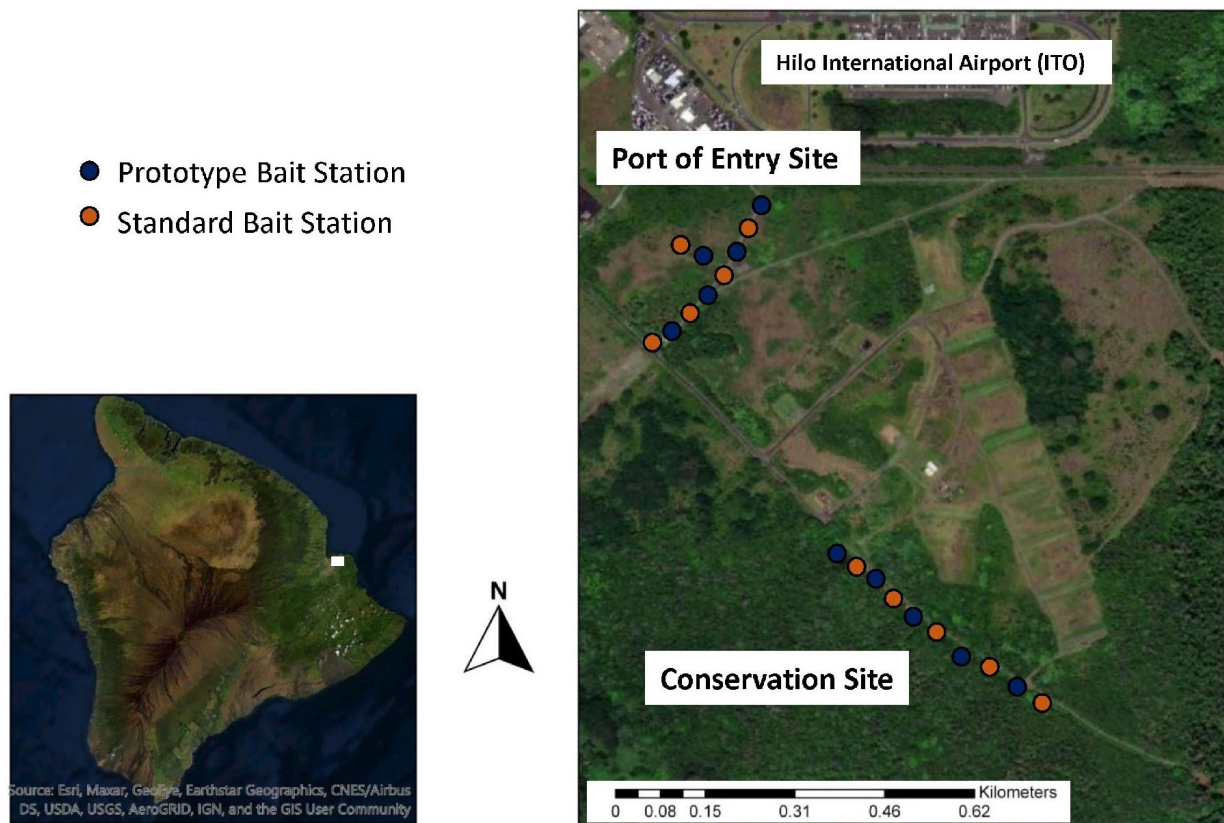
Each laboratory enclosure was randomly assigned either prototype A, B, C, or type D (Figure 1). All four types of bait stations were tested in each round. At the start of the test, the bait station in each enclosure was filled with two pieces (70 grams) of the non-toxic Fish-based Bait for Mongooses. Bait pieces were secured in the bait station's internal chamber on metal rods. Each bait station type was tested on four different mongooses (two males, two females, one mongoose per enclosure at a time) for a 24- or 48-hour exposure period. Four separate enclosures, each with a unique bait station type, allowed for four mongooses (one per enclosure) to be tested at a given time. Thus, there were four rounds of four mongooses during each

test (two males, two females per round). A total of 16 mongooses (8 males, 8 females) were randomly assigned to the four bait station types, stratified by sex, while ensuring that there was no size bias among bait station test groups. After each 24-hour period within the 24- to 48-hour total exposure period, the amount of test bait consumed, remaining, and spilled (found outside of the bait station) was collected in each enclosure and weighed. Only bait found outside of a bait station was considering spilled. Bait removed from the rod but remaining in the bait station's internal chamber was considered remaining bait (not spilled). Bait cached or stored by the mongoose within the enclosure was considered the same as spilled bait.

Due to daily fluctuating humidity levels in the animal testing room, baits were expected to gain or lose small amounts of moisture from the air each day in the period between when the bait was first offered and when the amount remaining the following day was weighed. Therefore, two separate samples of baits were weighed (approximately 70 grams) in identical plastic cups and placed in the same room as the test animals over the same time period each day. They were weighed the following day at the same time as the bait remaining in each enclosure was weighed. The amounts of bait offered each day were then corrected by multiplying a correction factor calculated as the final weight of the bait divided by the initial weight. The corrected amount offered/refilled the previous day was used to calculate the amount consumed by each mongoose from each bait station (i.e., amount eaten = corrected amount offered the previous day minus the amount remaining in bait station and spilled from the previous day). If bait within the bait station was accessed by mongooses and more than 50% was eaten before the end of first 24-hour period, the trial ended. If less than 50% was eaten, the trial continued for another 24-hour period (day 2). The uneaten and spilled/ cached bait was recovered, weighed, and then placed back on the metal rod in the bait station if the trial continued. Capturing mongooses inside the habitat was done by placing baited cage traps inside the enclosure. At the end of the test period, mongooses were humanely euthanized.

While on test, mongooses were observed via remote cameras with a live feed to another room to avoid animal disturbance for twice daily health checks (morning check 8:00 am–10:00 am, afternoon check 2:00 pm–4:00 pm). A motion-activated Hyperfire™ field camera (Reconyx®, Holmen, WI) was placed in each enclosure facing the bait station. Bait station discovery, feeding, and bait caching during the 12-hour active diurnal period were recorded.

Animal physical health was checked in person at least two times daily and recorded in the animal observation form. Water was checked and replenished daily. Each enclosure was completely cleaned between test groups and study subjects. Grasses, water bottles, refugia tubes, etc., were replaced for each test.

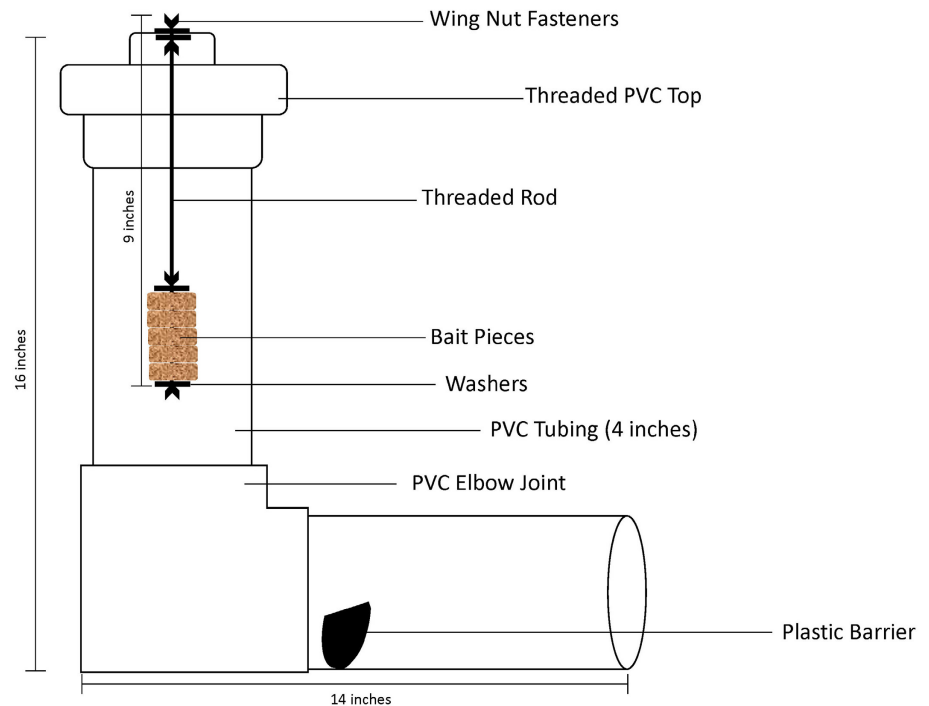


**Figure 2.** Map of field sites and bait station locations.

### *Field trials- free-ranging mongooses*

All field experiment activities occurred on two separate sites in Hawai'i that reflect typical areas managed for rodents and mongooses. These two areas were chosen to represent: 1) a forested “conservation area”, and 2) a “port of entry” along transportation routes near the airport. Both sites were within the Keaukaha Military Reservation, with the first site being in a forested area with dense vegetation and the second near the Hilo International Airport along roads and transport staging areas (Figure 2).

For the field trials, we tested the Protecta LP (type D) and a prototype design most similar to prototype C based on the laboratory trial results (Figure 3). Prototype C was selected due to its performance in the laboratory trials and its single-entry design, which may be less attractive to nontarget rodents in the field, as rodents are more likely to enter something with a second entrance/alternate escape route (Timm 1994). Modifications made after the laboratory trial included adding washers on the metal rod to secure the bait in place and adding an internal lip within the bottom PVC tube to minimize spillage outside of the bait station. Non-toxic Fish-based Bait for Mongooses test baits were secured on wire rods and placed in the prototype and commercial bait stations. The prototype design had vertical threaded rod placement with washers to secure the bait within the top of the PVC tube (Figure 3), while the Protecta LP had lateral rod placement without washers on the floor of the bait station (Figure S3 and S4). At each of the two



**Figure 3.** Schematic of the mongoose-targeted bait station prototype design tested for the delivery of Fish-based Bait for Mongooses made out of polyvinyl chloride (PVC) materials including a raised threaded bait rod secured with wing nut fasteners and washers and a plastic barrier to prevent spillage.

sites, five of each of the two bait station types were placed in an alternating fashion spaced at approximately 50 m, for a total of 10 of each type (Figure 2). Each bait station was provisioned with two bait pieces of ~ 35 g each. All bait stations were staked down or secured with rocks to reduce the likelihood of heavy rain or wind affecting or damaging stations before the study completion (Figures S3 and S4).

All bait stations per site were monitored by a Reconyx Hyperfire™ weather-proof, motion-sensor infra-red field camera (no-glow covert infrared) with 0.2 s trigger speed, to record bait interactions by mongoose and potential nontarget species (e.g., ground-foraging birds, rodents, and feral pigs). Cameras were deployed in the early morning and set to record motion-activated images in bursts of three images at 5-second intervals per event continuously throughout the study. During daylight, diurnal mongooses were typically recorded in color images, while nocturnal activity of mice and rats was typically recorded in grayscale images with infrared illumination. A visitation was recorded if the animal was recorded in the frame of the image. We recorded indices of all species that visited bait stations from these images.

Bait stations were also physically examined daily by field technicians in the mornings between 8:00 am and 12:30 pm. The bait remaining in bait stations and spilled outside of the bait stations were observed and recorded. Bait was also monitored for weathering, spoilage, and consumption by insects, mollusks, and other detritivores. Bait condition was evaluated and



camera data downloaded daily for the first five days, followed by an evaluation and download every three days thereafter until the bait was completely consumed at each site.

### *Statistical analyses*

For the laboratory enclosure trials, descriptive statistics (mean, standard deviation, and range) were calculated for total amount of bait consumption and total amount of bait spillage over the 1- or 2-day exposure period. The amount of bait eaten and spilled daily from each bait station was calculated as the weight offered (corrected for moisture loss/gain) minus the weight remaining and spilled. Differences in bait consumption and spillage by sex and bait station prototype were calculated with an analysis of variance (ANOVA).

For field trials, descriptive statistics were calculated for bait remaining in the bait station, bait spillage, and number of images of mongooses and other nontarget animals captured on the field cameras. Significant differences in bait consumption, spillage, and images by bait station type and site were calculated using *t*-tests. All procedures were performed in Microsoft Excel (Version 2302).

## **Results**

### *Laboratory enclosure trials*

Prior to the start of the trial, all 8 male and 8 female mongooses appeared normal. Mongoose body weights ranged from 368 g to 911 g upon arrival at the NWRC Hawai'i Field Station (Table S1).

Over the 1–2 day exposure periods, the total amount of bait eaten did not differ by bait station type, and ranged from 19.22 g to 64.05 g [ANOVA,  $F_{15} = 0.65$ ,  $p = 0.604$ ]. Male mongooses consumed more bait than females [ANOVA,  $F_{15} = 5.30$ ,  $p = 0.050$ ]. There was spillage of the bait, within and outside of the bait station, across individuals and all bait station types. Over the exposure periods, the total amount of bait spilled by bait station type is reported in Table 1 and plotted in Figure 4. Although, on average there was more spillage in prototypes A and D than with prototypes B and C, it was not statistically significant [ANOVA,  $F_{15} = 3.15$ ,  $p = 0.086$ ] (Table 1). Bait spillage did not significantly differ between sexes [ANOVA,  $F_{15} = 1.05$ ,  $p = 0.336$ ] (Table 1). Bait consumption and spillage results are plotted in Figure 4.

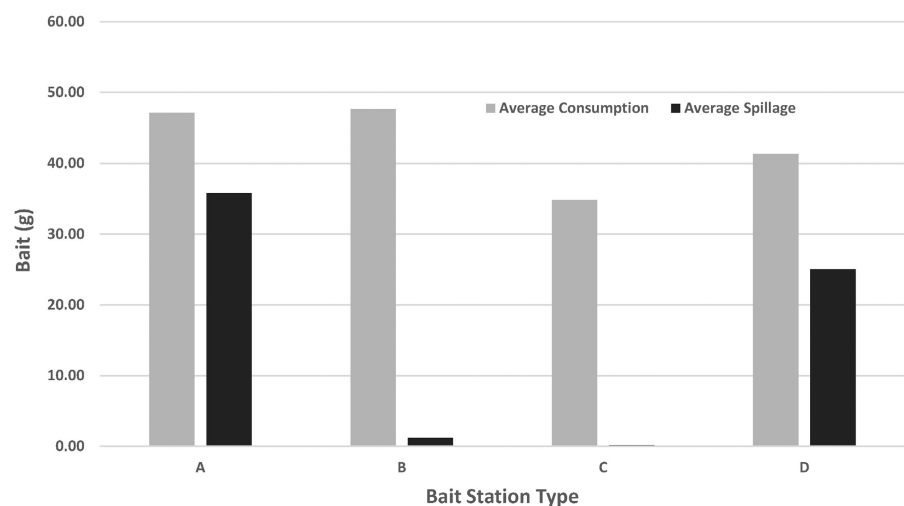
### *Field study*

#### Bait station visitations by mongooses

Camera images of mongooses were captured at all bait stations, for a total of 5,330 images (Table S2). Mongooses were confirmed to have accessed and consumed the bait at both bait station types tested. The earliest time recorded

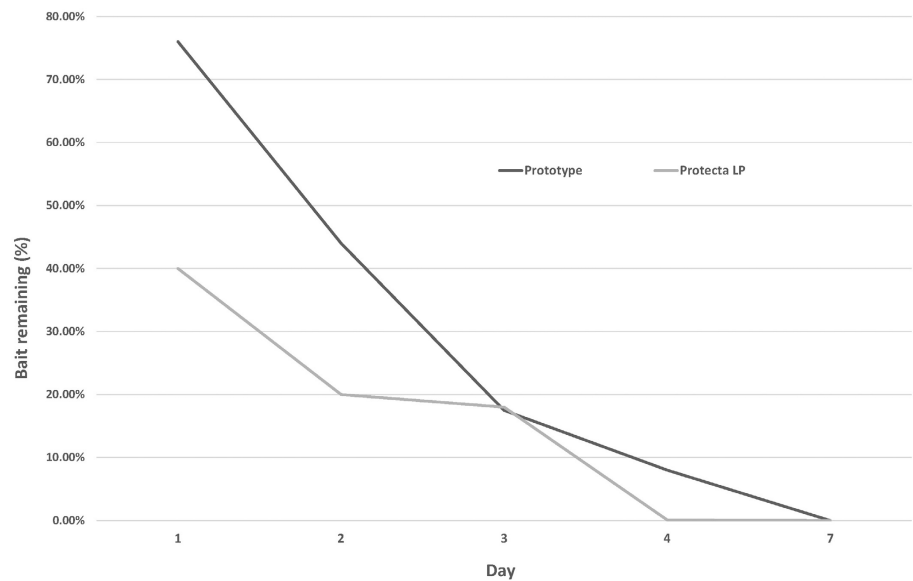
**Table 1.** Non-toxic Fish-based Bait for Mongooses bait consumption and spillage (total corrected) descriptive statistics and analysis of variance by bait station type and sex (ANOVA results) in laboratory study.

Consumption	Bait station type				Sex	
	A	B	C	D	Males	Females
mean (g)	47.14	47.64	36.55	41.30	50.66	35.65
SD	20.52	10.86	11.11	14.90	14.07	9.87
min (g)	19.22	34.01	22.14	21.35	37.81	19.22
max (g)	64.05	57.12	48.45	55.46	64.05	57.12
variance	421.41	118.03	117.52	222.10	97.45	198.02
df	15				15	
f	0.65				5.30	
p-value	0.604				0.050	
<b>Spillage</b>						
mean (g)	35.82	1.18	0.16	25.09	10.44	20.68
SD	29.67	1.32	0.32	22.36	16.57	28.25
min (g)	3.75	0.00	0.00	12.10	0.00	0.00
max (g)	70.15	3.04	0.64	58.53	49.38	70.15
variance	880.39	1.74	0.10	499.89	274.42	798.20
df	15				15	
f	3.15				1.05	
p-value	0.086				0.336	



**Figure 4.** Average amount of non-toxic Fish-based Bait for Mongooses consumed (g) and spilled (total corrected) per individual from each bait station type (A, B, C, and D) by mongooses in the laboratory study.

on mongooses accessing the bait stations was 4:29 am and the latest image recorded was at 8:16 pm (Figure S5). The mean time until mongooses were first detected at a bait station was 18:07 (hh:mm) after placement (SD = 19.05, range 00:14–68:52; Table S2). This was not different between bait station types [t-test,  $T_{18} = 0.09$ ,  $p = 0.46$ ] but differed between the “Conservation Area” site and “Port of Entry” site [t-test,  $T_{18} = -2.17$ ,  $p = 0.022$ ; Table S3]. There were more mongoose images captured at the prototype bait stations than at the Protecta LP bait stations [t-test,  $T_{18} = 2.54$ ,  $p = 0.010$ ]. Number of mongooses images did not vary significantly between sites [t-test,  $T_{18} = 0.01$ ,  $p = 0.495$ ; Table S3].



**Figure 5.** Mean amount of non-toxic Fish-based Bait for Mongooses remaining per day (%) from each bait station by bait station type (modified prototype C and standard Protecta® LP bait station) in the field study.

### Bait consumption

All bait consumption in the field was attributed to mongooses, based on camera images. The amount of bait remaining by day over the test periods is plotted in Figure 5. Across all bait stations, the mean number of days until bait was completely consumed was 2.7 days (SD = 1.81, range 1–7). All bait was completely consumed after three days in the “Conservation Area” site and after 7 days in the “Port of Entry” site. On average, the bait remained in the “Port of Entry” site bait stations for longer (M = 3.7 days, SD = 2.0, range 1–7) than those in the “Conservation Area” site (M = 1.7 days, SD = 0.8, range 1–3) [t-test,  $T_{18} = 2.92$ ,  $p = 0.005$ ; Table S4]. On average, the bait remained in the prototype bait stations longer (M = 3.2 days, SD = 1.61, range 1–7) than in the Protecta LP bait stations (M = 2.2 days, SD = 1.93, range 1–7), but this difference was not significant [t-test,  $T_{18} = 1.25$ ,  $p = 0.113$ ; Table S4].

### Bait spillage

All bait spillage in the field study was attributed to mongooses. Average spillage was less than 1% for both bait station types (Table S5). Bait spillage included crumbs of bait inside the bait station and outside the bait station. However, the majority of spillage found outside of the bait station were the discarded biodegradable bait skins/wrappers that encase the fish-based sausage bait, which do not contain diphacinone in the toxic bait version, and were considered to be < 1% spillage (Figure S6). The bait spillage was not different between prototype and Protecta LP bait stations [t-test,  $T_{88} = -1.12$ ,  $p = 0.133$ ] or between sites [t-test,  $T_{88} = -0.58$ ,  $p = 0.281$ ; Table S5].

**Table 2.** Nontarget species recorded on game cameras and their interactions at bait stations during the field study.

Species	Bait station images	Accessed bait station	Sniffed or ignored
Polynesian rat ( <i>Rattus exulans</i> )	38	1	37
Feral goat ( <i>Capra hircus</i> )	17	0	17
Feral swine ( <i>Sus scrofa</i> )	11	0	11
Common myna ( <i>Acridotheres tristis</i> )	4	0	4
Feral chicken ( <i>Gallus gallus</i> )	3	0	3
Feral domestic cat ( <i>Felis catus</i> )	1	0	1

### Nontarget activity at bait stations

Images of nontarget species were captured at 50% of the prototype bait stations (37 images) and 70% of the standard commercial bait stations (37 images) (Table S2). Of those nontarget images, all were species that are non-native or introduced to Hawai'i: 38 were Polynesian rats (*Rattus exulans*), 17 were feral goats (*Capra hircus*), 11 were feral swine (*Sus scrofa*), 4 were Common mynas (*Acridotheres tristis*), 3 were feral chickens (*Gallus gallus*), and 1 was a feral domestic cat (*Felis catus*) (Table 2). These images showed none of these animals accessing the bait stations, only passing by, sniffing the outside, etc., with the exception of one rat recorded entering a Protecta LP bait station. Upon visual inspection the next morning, the bait appeared to be intact, and showed no evidence of consumption by the rodent (Figure S7). The number of images of nontarget species ( $M = 3.70$ ,  $SD = 4.61$ , range 0–17) was significantly less than the number of images of mongooses ( $M = 263.30$ ,  $SD = 218.44$ , range 19–648) [t-test,  $T_{19} = 6.09$ ,  $p < 0.0001$ ; Table S3]. The number of nontarget images did not differ by bait station type [t-test,  $T_{18} = 0$ ,  $p = 0.500$ ] or site [t-test,  $T_{18} = -0.09$ ,  $p = 0.463$ ; Table S3].

During visual bait observations, slugs were present in 30% of the prototype bait stations and 70% of the commercial bait stations. Ants were present in 10% of the prototype bait stations and 30% of the commercial bait stations. However, the slugs and ants were never observed on the actual bait and the condition of the bait remained intact in the presence of invertebrates, without signs of degradation (Figure S8).

### **Discussion**

Mongoose freely utilized bait stations and consumed non-toxic Fish-based Bait for Mongooses from all three prototypes and the commercial Protecta LP rodenticide bait stations. In the laboratory trials, mongooses spilled less bait from Prototypes B and C. We hypothesize that the PVC tube designs created a suitable environment for mongooses to eat the bait in place, and if it spilled, the bait remained within the bait station due to the design, reducing the amount of spillage outside of the bait station. We selected a modified version of Prototype C (Figure 3) for the field trial because we reasoned that the single-entry design may be less attractive to

nontarget rodents in the field. Most commercially available rodenticide bait stations have two entry holes because rodents are more likely to enter a confined area with an alternate escape route (Timm 1994).

The material cost to construct the prototype bait station was approximately \$21.59 each, while the cost of the Protecta LP bait station at \$14.48 each at the time of purchase. Due to the simple design of the prototype, a technician can easily fabricate ten bait stations in an hour. If you divide by the hourly cost of a technician (\$15/hour), each bait station's labor cost adds only \$1.50 to the total cost.

The field trials validated the performance of this prototype PVC bait station design as similar to the commercial tamper-resistant Protecta LP rat bait station, a bait station that is readily available to land managers and currently used with registered rodenticides. Free-ranging mongooses easily accessed both Protecta LP and prototype bait stations in the field and quickly found and consumed the non-toxic fish-based bait. Bait was consumed within 1 to 7 days at the study sites, even with an abundance of diverse alternate food resources in the habitat (Pitt et al. 2015). Bait persisted in the field longer in the prototype PVC bait stations (mean of 3.2 days), compared to the standard commercial bait stations (mean of 2.2 days), before being consumed completely. This is potentially due to the design of the prototype bait stations to secure the bait to the wire rod with washers, in an attempt to avoid mongooses removing and caching the entire sausage-like bait piece. Caching bait decreases availability in the bait station to other conspecifics and increases the probability of consumption by a nontarget outside of the bait station (Thomson and Kok 2002; Brakes and Smith 2005). To ensure that each mongoose consumes a lethal dose of diphacinone, prolonged bait availability is advantageous. Bait consumption rates suggest that bait may need to be replenished at least every week to maintain continuous feedings in areas of high mongoose density.

Based on the number of images, there was significantly more mongoose activity at prototype bait stations than at Protecta LP bait stations. This is likely due to repeat visitation at prototype bait stations by mongooses for multiple feedings. There was minimal spillage at both types of bait stations, which mostly consisted of discarded nontoxic biodegradable bait skins/wrappers. While we were not able to determine bait removal and transport outside of bait stations, it is presumed that the majority of bait was consumed inside the bait station, as the bait was secured with a metal rod and washers in prototype bait stations. Although, there were some images of mongooses holding bait/wrappers in their mouth outside of bait stations, potentially for caching at their dens or consuming later.

Most of the camera images recorded mongoose foraging at bait stations during daylight hours, but some were crepuscular. Our mongoose foraging times were earlier and later than previous findings (Pitt et al. 2015). It was not uncommon to see multiple mongooses in a camera image (Figure S9)

or mongooses sitting outside of bait stations for an extended duration, potentially protecting the bait station food source from cohorts.

There was no evidence of nontarget species accessing bait inside or spilled bait outside of the prototype bait stations. The only observation of rodent access was by one individual rat entering a Protecta LP bait station. There was no evidence of bait consumption by nontarget animals at either bait station type. Overall, the prototype bait station and commercial tamper-resistant design were equally effective at targeting mongooses and allowing them to consume the non-toxic Fish-based Bait for Mongooses.

Bait condition throughout the trial appeared to be unaltered with no evidence of mold, degradation, or consumption by arthropods. There were some slugs and ants present at bait stations, but they did not appear to consume or degrade the bait. The prototype bait stations appeared to be less accessible than the commercial rat bait stations to slugs and ants.

This work is part of a larger project to develop and register a novel vertebrate toxicant and delivery system to control the invasive small Indian mongoose in the U.S. (Antaky et al. 2023). An application for an EUP has been approved by the EPA for a larger field efficacy and product performance study (EUP No.: 56228-EUP-45) which will support a subsequent national registration application. Based on this research, the prototype PVC bait station will be selected for use in the product performance (field) trials under an Experimental Use Permit for the registration of the Fish-based Bait for Mongooses. The tamper-resistant commercial Protecta LP bait station also performed well and may still be used in specific locations when required by EPA to reduce risks to children and pets. Deployment modifications may be needed, and the use of bait stations may be curtailed in habitats with high feral swine density or other large mammals that might destroy or interfere with the bait stations. The forthcoming EPA registration of Fish-based Bait for Mongooses for use in bait stations will allow delivery in novel bait stations or in commercially available rodenticide bait stations that meet the construction requirements. Building on the promising results from past research (Antaky et al. 2023), these studies continue the momentum toward field deployment of a new effective toxic bait for mongoose management and control in agriculture, biosecurity, conservation, and human health applications.

## Conclusion

This research facilitated the development and testing of a novel bait station design for future delivery of toxic baits to invasive mongooses, with the aims of minimize nontarget take while remaining cost-effective and easy to use. The laboratory study examined a suite of four bait station designs in captive enclosures to evaluate bait accessibility by mongooses and to minimize bait spillage. The field study examined one of the best performing

prototype bait stations, as identified in the laboratory, and compared a commercially available rat bait station by measuring bait uptake by mongooses and potential nontarget interactions in the field. We found that both the “L” PVC tube design with a single single-entry point and Protecta LP commercial rodenticide bait station worked well in the laboratory and field to minimize spillage by mongooses and nontarget risk.

Our testing suggests that the PVC prototype bait station can provide an effective alternative to commercial rodent bait stations to control the invasive small Indian mongoose in Hawai‘i and globally. Further research is needed to test the prototype bait station over longer periods of time and across different environments, optimize baiting strategies, and identify bait deployment schedules to meet management objectives for a given area. Managers experiencing higher nontarget interference rates than we observed at our study sites may wish to evaluate the utility of our novel prototype alternatives for their particular challenges.

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### Authors’ contribution

C.A. research conceptualization, sample design and methodology, investigation and data collection, data analysis and interpretation, writing; R.S. research conceptualization, sample design and methodology, investigation and data collection, data analysis and interpretation, and writing; I.L. sample design and methodology, investigation and data collection, editing; S.S. research conceptualization, sample design and methodology, review and editing; E.R. research conceptualization, review and editing. S.H. research conceptualization, sample design and methodology, review and editing.

### Ethics and permits

Research was conducted under IACUC Protocols QA-3388 and QA-3439 and ESA Informal Consultation #01EPIF00-2022-I-0022 and following the State of Hawaii Department of Agriculture permit (HI-ISL-2014-01).

### References

- Abernethy EF, Turner KL, Beasley JC, DeVault TL, Pitt WC, Rhodes Jr. OE (2016) Carcasses of invasive species are predominantly utilized by invasive scavengers in an island ecosystem. *Ecosphere* 7: e01496, <https://doi.org/10.1002/ecs2.1496>
- Antaky CC, Hess SC, Ruell EW, Leinbach IL, Siers SR, Sugihara RT (2023) The path to U.S. national registration of a toxic bait for the control of the small Indian mongoose. *Human-Wildlife Interactions* (*in press*)

- Alterio N, Brown K, Moller H (1997) Secondary poisoning of mustelids in a New Zealand *Nothofagus* forest. *Journal of Zoology* 243: 863–869, <https://doi.org/10.1111/j.1469-7998.1997.tb01986.x>
- Bailey EP (1993) Introduction of foxes to Alaskan islands-history, effects on avifauna, and eradication. Fish and Wildlife Service, Homer AK, 62 pp
- Barun A, Hanson CC, Campbell KJ, Simberloff D (2011) A review of small Indian mongoose management and eradications on islands. In: Veitch CR, Clout MN, Towns DR (eds), *Island invasives: eradication and management*. IUCN, Gland, Switzerland, pp 17–25
- Berentsen AR, Pitt WC, Sugihara RT (2018) Ecology of the small Indian mongoose (*Herpestes auropunctatus*) in North America. In: Pitt WC, Beasley JC, Witmer GW (eds), *Ecology and management of terrestrial vertebrate invasive species in the United States*. CRC Press, Boca Raton, Florida, USA, pp 251–267, <https://doi.org/10.1201/9781315157078-12>
- Brakes CR, Smith RH (2005) Exposure of non-target small mammals to rodenticides: short-term effects, recovery and implications for secondary poisoning. *Journal of Applied Ecology* 42: 118–128, <https://doi.org/10.1111/j.1365-2664.2005.00997.x>
- Everard COR, Green AE, Glosser JW (1976) Leptospirosis in Trinidad and Grenada, with special reference to the mongoose. *Royal Society of Tropical Medicine and Hygiene* 70: 57–61, [https://doi.org/10.1016/0035-9203\(76\)90008-0](https://doi.org/10.1016/0035-9203(76)90008-0)
- Hays WS, Conant S (2007) Biology and impacts of Pacific Island invasive species. 1. A worldwide review of effects of the small Indian mongoose, *Herpestes javanicus* (Carnivora: Herpestidae). *Pacific Science* 61: 3–16, <https://doi.org/10.1353/psc.2007.0006>
- Howald G, Donlan CJ, Galván JP, Russell JC, Parkes J, Samaniego A, Wang Y, Veitch D, Genovesi P, Pascal M, Saunders A (2007) Invasive rodent eradication on islands. *Conservation Biology* 2: 1258–1268, <https://doi.org/10.1111/j.1523-1739.2007.00755.x>
- Keith JO, Hirata DN, Epsy DL, Greiner S, Griffin D (1989) Field Evaluation of 0.00025% Diphacinone bait for mongoose control in Hawaii. DWRC Job Comp. Rpt., Denver, CO, 35 pp
- Nellis DW, Everard CO (1983) The biology of the mongoose in the Caribbean. *Studies on the fauna of Curacao and other Caribbean Islands* 64(1): 1–62
- Phillips RB, Lucey B (2016) Kauai mongoose standard operating procedures to conduct and island-wide status assessment and early detection rapid response. US Fish and Wildlife Service, Pacific Islands Fish and Wildlife Office. Unpublished report, 25 pp
- Pitt WC, Sugihara RT, Berentsen AR (2015) Effect of travel distance, home range, and bait on the management of small Indian mongooses, *Herpestes auropunctatus*. *Biological Invasions* 17: 1743–1759, <https://doi.org/10.1007/s10530-014-0831-x>
- Roerk LS, Nietmann L, Works AJ (2022) Efficiency and Efficacy of DOC-200 Versus Tomahawk Traps for Controlling Small Indian Mongoose, *Herpestes auropunctatus* (Carnivora: Herpestidae) in Wetland Wildlife Sanctuaries. *Pacific Science* 76: 201–207, <https://doi.org/10.2984/76.2.8>
- Ruell EW, Niebuhr CN, Sugihara RT, Siers SR (2019) An evaluation of the registration and use prospects for four candidate toxicants for controlling invasive mongooses (*Herpestes javanicus auropunctatus*). *Management of Biological Invasions* 10: 573–596, <https://doi.org/10.3391/mbi.2019.10.3.11>
- Seetahal JFR, Vokaty A, Vigilato MAN, Carrington CVF, Pradel J, Louison B., Sauers AV, Roopnarine R, Arrebatto JCG, Millien MF, James C, Rupprecht CE (2018) Rabies in the Caribbean: A Situational Analysis and Historic Review. *Tropical Medicine and Infectious Diseases* 3: 89, <https://doi.org/10.3390/tropicalmed3030089>
- Siers SR, Sugihara RT, Ruell EW, Leinbach IL, Sedgwick D, Niebuhr CN (2020) Development and testing of a matrix for mongoose toxic bait: nontoxic bait acceptance cage trials. Final Report QA-2832. USDA, APHIS, WS, NWRC. Hilo, HI, 13 pp
- Sugihara RT, Pitt WC, Berentsen AR, Payne CG (2018) Evaluation of the palatability and toxicity of candidate baits and toxicants for mongooses (*Herpestes auropunctatus*). *European Journal of Wildlife Research* 64: 1–9. <https://doi.org/10.1007/s10344-017-1163-9>
- Thomson PC, Kok NE (2002) The fate of dried meat baits laid for fox control: the effects of bait presentation on take by foxes and non-target species, and on caching by foxes. *Wildlife Research* 29: 371–377, <https://doi.org/10.1071/WR01098>
- Timm RM (1994) Norway rats. *The Handbook: Prevention and Control of Wildlife Damage*, University of Nebraska, Lincoln, 5 pp
- Underwood JG, Silbernagle M, Nishimoto M, Uychara K (2013) Managing conservation reliant species: Hawaii's endangered endemic waterbirds. *PLoS ONE* 8: e67872, <https://doi.org/10.1371/journal.pone.0067872>
- Wong M, Katz AR, Li D, Wilcox BA (2012) Leptospira infection prevalence in small mammal host populations on three Hawaiian Islands. *American Journal of Tropical Medicine and Hygiene* 87: 337–341, <https://doi.org/10.4269/ajtmh.2012.12-0187>



- Yamada F, Sugimura K (2004) Negative impact of an invasive small Indian mongoose *Herpestes javanicus* on native wildlife species and evaluation of a control project in Amami-Oshima and Okinawa Islands, Japan. *Global Environmental Research* 8(2): 117–124
- Young LC, VanderWerf EA, Mitchell C, Yeun E, Miller CJ, Smith DG, Swenson C (2012) The use of predator proof fencing as a management tool in the Hawaiian Islands: a case study of Kaena Point Natural Area Reserve. Technical Report No. 180. Pacific Cooperative Studies Unit, University of Hawai'i, Honolulu, Hawai'i, 87 pp

### Supplementary material

The following supplementary material is available for this article:

**Table S1.** Mongoose body weights on the day of arrival in the laboratory study.

**Table S2.** Analysis of field camera images of mongoose and nontargets at bait stations and descriptive statistics in the field study.

**Table S3.** *T*-test results of mongoose and nontargets at bait stations in the field study.

**Table S4.** *T*-test results of bait consumption in the field study.

**Table S5.** *T*-test results of bait spillage in the field study.

**Figure S1.** Two pieces of “Fish-based Bait for Mongooses”.

**Figure S2.** Artificial test enclosures staged at the NWRC Hawaii Field Station.

**Figure S3.** Prototype Bait Station deployed at the field sites.

**Figure S4.** Tamper-resistant commercial bait station deployed at the field sites.

**Figure S5.** Images of earliest and latest recorded mongoose bait station visitation times.

**Figure S6.** Images of spillage and bait wrappers at the field sites.

**Figure S7.** Images of a rat entering the bait station and of the bait the following day.

**Figure S8.** Images of invertebrate presence in bait stations.

**Figure S9.** Game camera image of mongoose activity at bait stations in the field study.

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