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Pilot study for invasive brown treesnake baiting in residential areas

Shane R. Siers, USDA, APHIS, Wildlife Services, National Wildlife Research Center, 233 Pangelinan Way, Barrigada, Guam 96913, USA *Shane.r.siers@usda.gov*

Juan-Carlos Mungaray, Research Corporation of the University of Guam, Dean's Circle House #24, Mangilao, Guam 96913, USA

Jordan I. S. Barcinas, Research Corporation of the University of Guam, Dean's Circle House #24, Mangilao, Guam 96913, USA

Jeried E. Calaor, Research Corporation of the University of Guam, Dean's Circle House #24, Mangilao, Guam 96913, USA

Rachel M. Volsteadt, USDA, APHIS, Wildlife Services, National Wildlife Research Center, 233 Pangelinan Way, Barrigada, Guam 96913, USA

Martin Kastner, Iowa State University, Department of Ecology, Evolution and Organismal Biology, 251 Bessey Hall, Ames, IA 50011, USA

Scott M. Goetz, U.S. Geological Survey, Pacific Island Ecosystems Research Center, Hawai'i National Park, HI 96718, USA

Melia G. Nafus, U.S. Geological Survey, Pacific Island Ecosystems Research Center, Hawai'i National Park, HI 96718, USA

Thomas C. Hall, USDA, APHIS, Wildlife Services, Operational Support Staff, 2150 Centre Avenue, Building B, Fort Collins, CO 80526, USA

Abstract: The nocturnal brown treesnake (*Boiga irregularis*; BTS) was accidentally introduced to the island of Guam, USA, in the Western Pacific in 1945. The BTS has spread throughout all terrestrial habitats, causing wildlife loss and economic damage. Several tools and techniques have been developed to locally reduce BTS numbers and prevent their spread to other islands. The common analgesic acetaminophen has been registered as a low-risk pesticide to manage BTS in non-residential areas. Prior to a more intensive toxic baiting campaign on Andersen Air Force Base, Guam, as part of a larger study to evaluate the effects of BTS control on native bird survival, we conducted a pilot study from May 18 to October 6, 2020, to assess the efficacy and safety of BTS baiting within a residential area. We administered large and small domesticated mouse (*Mus musculus*) and bird chick (*Coturnix japonica* and *Gallus domesticus*) nontoxic carrion baits (without acetaminophen) in bait stations to evaluate BTS and nontarget bait take rates, bait preferences, movements following nighttime bait uptakes, and interactions of humans and domestic animals with baits. We monitored baits with cameras and implanted them with radio-transmitters to verify the species taking the bait and to track BTS to their daytime sheltering locations. Successful BTS bait take rates were low at 20 of 482 baits (4.1%), as were nontarget bait take rates (4 baits; 0.8%). No preference among bait types was discernible, though power to detect differences was limited due to low overall uptake. All first daytime refugia were within vegetation except for 1 location on the roof of a house. No evidence of bait removal or human tampering was found at any of the bait stations. Based on our pilot study, there appeared to be little human or nontarget risk from acetaminophen baiting in this relatively uniform and sparsely vegetated residential area. Because most of the residential areas on Guam are much more variable, similar assessments within a broader diversity of residential areas may be advisable before promoting large-scale use of acetaminophen for managing BTS in close contact with human habitations.

Key words: acetaminophen, bait preference, *Boiga irregularis*, brown treesnake, conservation, Guam, interdiction, nontarget species, quality of life, risk assessment, toxic baiting, vertebrate pesticide

The nocturnal, arboreal brown treesnake (*Boiga irregularis*; BTS) was accidentally introduced to the island of Guam, USA, in the Western Pacific, most likely in shipments of military equipment and salvage after the end of World War II in 1945 (Rodda and Savidge 2007, Richmond et al. 2014). By the 1980s, their spread throughout all terrestrial habitats on the island coincided with a wave of wildlife loss and economic damage (Savidge 1987, Fritts and Rodda 1998, Fritts 2002, Wiles et al. 2003, Rodda and Savidge 2007). Since the 1990s, several tools and

Figure 1. Brown treesnake (*Boiga irregularis*) discovered by U.S. Department of Agriculture (USDA) Wildlife Services during an outgoing household goods shipment from Andersen Air Force Base, Guam, USA (*photo courtesy of USDA*).

techniques have been developed to locally reduce BTS numbers and prevent their spread to other islands (Clark et al. 2018).

The human pharmaceutical acetaminophen was discovered to be an effective oral toxicant for lethal control of BTS with a relatively low environmental risk profile (Savarie et al. 2000, Johnston et al. 2002, Mathies and Mauldin 2020). The U.S. Department of Agriculture Wildlife Services (WS) subsequently registered a tablet of 80 mg acetaminophen (N-acetyl-para-aminophenol, paracetamol) as a pesticide with the U.S. Environmental Protection Agency (EPA) under the product name "Acetaminophen for Brown Treesnake Control" (EPA Registration No. 56228-34). Toxic baiting of BTS using the WS-registered acetaminophen tablet, which are typically inserted within a dead neonatal mouse bait (DNM; *Mus musculus*; approximately 5 g), has since become a cost-effective mainstay of BTS management and interdiction operations (Clark et al. 2012, 2018). These acetaminophen mouse baits (AMB) are typically placed within plastic tube bait stations to prevent accidental exposure or bait interference by nontarget species such as birds, rats, crabs, and monitor lizards (*Varanus tsukamotoi*; Mathies et al. 2011, Lardner et al. 2013). Operational BTS management with AMB generally occurs around commercial and military air and sea ports to prevent accidental transport of BTS, around the perimeters of military bases, and around critical electrical infrastructure to prevent snake-caused power outages (Fritts 2002). Such AMB also have been adapted for aerial delivery via helicopter for landscape-scale BTS suppression in forests, in hopes of promoting wildlife recovery (Clark and Savarie 2012; Dorr et al. 2016; Siers et al. 2019*a*, 2020*a, b*).

Baiting with a toxicant for BTS has not previously occurred within close proximity to human habitations. In residential areas, BTS primarily pose a "quality of life" issue for humans and also can bring about inconvenience and economic harm by causing power outages (Fritts 2002). The BTS is rear-fanged and mildly venomous to mammals (Mackessy et al. 2006), but bites can be medically significant, especially to infants (Fritts et al. 1990, 1994). Most bites are defensive or feigned without fang engagement, but snakebites are often traumatic, particularly to children and their parents (Rodda and Savidge 2007).

The BTS has remarkable climbing abilities (Jayne and Riley 2007, Savidge et al. 2021), and nighttime "home invasions" by BTS are common and upsetting (Fritts 1988, Fritts et al. 1994). In addition to these unpleasant consequences of BTS in homes and yards, BTS also have been known to prey on pets (especially newborn dogs [*Canis familiaris*] and cage birds) and have negatively impacted production of poultry and eggs (Fritts and McCoid 1991, Rodda and Savidge 2007).

Guam also supports 2 military installations (Andersen Air Force Base [AAFB] and Naval Base Guam [NBG]), with a third (Base Camp Blaz) scheduled to house relocated U.S. Marines starting in 2025. Both AAFB and NBG support service members and families in residential areas within the boundaries of the installations. There is annual turnover of personnel and associated relocation of household goods to new duty stations, including Hawaiʻi, USA, an area of high invasion risk and great potential for ecological and economic catastrophe should such an invasion occur (Stanford and Rodda 2007, Shwiff et al. 2010, Burnett et al. 2012).

Funding is provided by the U.S. Navy to support the BTS interdiction program on the installations to prevent the accidental spread of the snake; interdiction is focused around ports of exit, cargo processing areas, and residential areas. Stowaway BTS have been found in shipments of household goods and vehicles originating from residential areas on Guam (Stanford and Rodda 2007, Perry and Vice 2009, Pitt et al. 2010; Figure 1).

Figure 2. This study evaluated the potential human and nontarget wildlife exposure to toxic baits for the removal of brown treesnakes (*Boiga irregularis*) from residential areas on Andersen Air Force Base, Guam, USA, for the protection of såli (Micronesian starling; Aplonis opaca), pictured. This pilot study using nontoxic baits occurred from May 18 to October 6, 2020 (*photo courtesy of M. Kastner*).

Ecologically, since the collapse of bird and rodent prey in Guam's forests (Savidge 1988, Wiles et al. 2003, Wiewel et al. 2009), the largest BTS tend to be found in residential areas where introduced birds and commensal rodents are more abundant (Savidge 1988, 1991; Siers 2015; Siers et al. 2017*a, b*), likely fueling increased reproductive output. Brown treesnakes in urban, peri-urban, and residential areas of Guam help maintain populations of the snake throughout the island (Savidge 1988, Siers 2015), although snakes are typically at lower abundances than in native jungle growth (Hall 1996). The high numbers of BTS on Guam were broadly publicized in the 1990s and had a minor impact on tourism (Hall 1996). Thus, BTS suppression in urban areas could reduce populations in surrounding environs and diminish the perception of potential tourists that Guam is "crawling with snakes."

Micronesian starlings (*Aplonis opaca*), or såli, the native CHamoru name, are 1 of only 2 native forest birds for which BTS predation did not result in extirpation or extinction on Guam (Savidge 1987, Wiles et al. 2003; Figure 2). However, their nesting and roosting range on Guam has contracted almost entirely to the urban and residential areas on Andersen Air Force Base (AAFB) that provide greater refugia from predation (Wiles et al. 1995, Pollock et al. 2021). While the såli population has partially recovered on AAFB (Pollock et al. 2021), they still suffer very high rates of predation by BTS and reduced recruitment rates (Wagner et al. 2018, Pollock et al. 2019).

In 2019, the U.S. Navy funded a multi-entity study to evaluate whether BTS management within their range could result in improved såli survival and to inform plans for future efforts to recover other native species. Few environments on Guam offer the opportunity to improve understanding of how BTS management should be implemented in bird-rich landscapes to maximize snake removal.

One of the initial stages of this project was to attempt to reduce BTS numbers by acetaminophen baiting in residential and other urban såli habitat. Nafus et al. (2021) indicated that individual BTS that have been successful at preying on birds tend to prefer bird-based baits or lures over rodents. It also has been speculated by BTS researchers that the traditional 5 g AMB might not be as attractive to larger BTS that prey on live birds and that larger baits could be more suitable for targeting these larger predators.

When BTS take a meal, they tend to become inactive for 5–7 days during digestion (Siers et al. 2018). Therefore, the location of a snake after feeding is of interest, particularly if that refugium is within household goods or vehicles that could be packed for shipment off-island. The use of the WS-registered acetaminophen tablet for BTS control in urban areas has been approved by the EPA and the Government of Guam (GovGuam) for use by federal, state, and territorial employees or persons under their direct supervision. This pesticide cannot be used in a manner where baits are accessible to children and domestic animals. Additionally, any new usage pattern for a pesticide warrants a cautious approach to ensure that undue risks are not posed to nontarget species, including humans and domestic animals. Wildlife Services is currently completing a risk assessment for acetaminophen bait use on Guam, as well as potentially for other islands where BTS may be found in the future.

In this phase of the overall study, we sought to characterize human and nontarget risks of baiting in residential areas on AAFB by implementing a pilot baiting regimen using nontoxic baits with the following research objectives:

evaluate rates of bait take by BTS and nontarget animals; evaluate population-level BTS preferences for bait size and type (rodent or bird); collect morphometric data on BTS taking baits and track them to their first refuge locations; and assess risks to humans and domestic animals from acetaminophen baiting for BTS control in residential areas.

Study area

Andersen Air Force Base is situated atop a limestone plateau in northern Guam at approximately 135–180 m above sea level (Figure 3). This study was conducted throughout 240 ha of the AAFB housing area and surrounding environs, centered at 13.5608° latitude and 144.9223° longitude (World Geodetic System 1984 datum), comprising primarily small residential structures and associated infrastructure such as parks, playgrounds, school, barracks, and commissary. Vegetation consists of sparse ornamental plants including the shade trees fish poison tree (*Barringtonia asiatica*), mastwood (*Calophyllum inophyllum*), ironwood (*Casuarina equisetifolia*), coconut (*Cocos nucifera*), flame tree (*Delonix regia*), banyan (*Ficus* spp.), noni (*Morinda citrifolia*), plumeria (*Plumeria rubra*), *Premna serratifolia* (no common name), pink trumpet tree (*Tabebuia heterophylla*), and molave (*Vitex parviflora*). The grounds are frequently maintained by a landscaping contractor. Såli use the area extensively for nesting and roosting; Savidge et al. (2018) set up 60 cedar or plastic board nest boxes mounted on structures resistant to climbing by BTS, and these boxes have been readily adopted by såli (Savidge et al. 2018, Pollock et al. 2021).

Wildlife Services conducts BTS management at strategic locations on Guam, including at AAFB, to minimize the potential for BTS to spread to other islands. The primary area of concern for WS is the flightline area of operations where aircraft and cargo depart from AAFB, on which a BTS could stow away. Wildlife Services operates and maintains a perimeter of AMB in bait stations, live-capture funnel traps containing live mouse lures, and conducts spotlighting (nighttime visual searching and hand capture) on perimeter fences (Vice et al. 2005; Clark et al. 2012, 2018) along the forest edges of the base, but not within the housing area.

From 2016 to 2020, WS operations on AAFB annually averaged the removal of 2,111 BTS with traps, 441 by spotlighting, and 1 with detector dogs (detector dogs capture few snakes, but these are mostly in very high-risk outbound cargo areas). Additionally, 9,133 AMB were removed from bait tubes annually; although some of these baits may not have been taken by snakes, nontarget take rates are typically quite low (Siers et al. 2018, 2019*b*, 2020*a*) and the vast majority likely resulted in the death of a BTS. These figures demon-

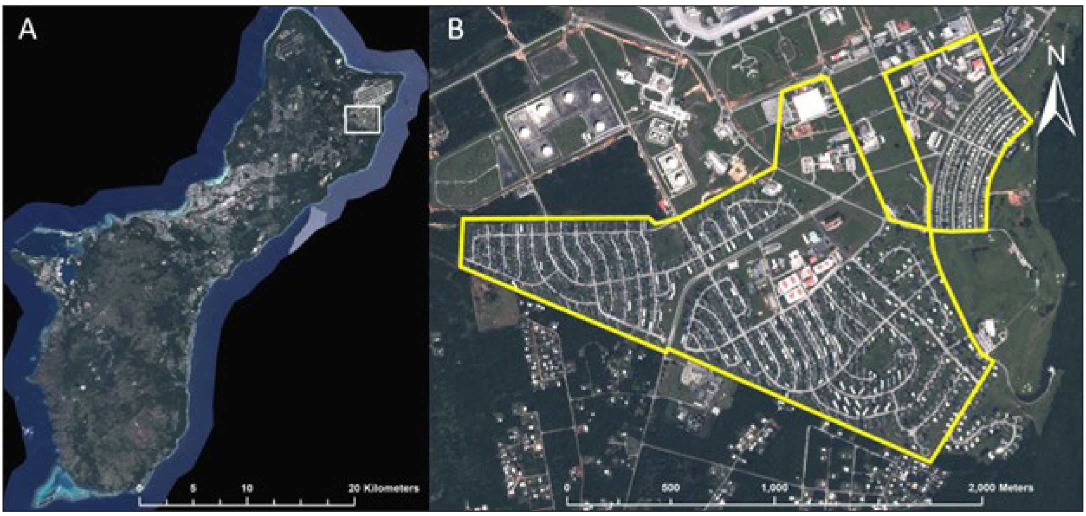

Figure 3. Location of Andersen Air Force Base (Guam, USA) residential housing and associated structures (barracks, schools, commissary, etc.), where this brown treesnake (*Boiga irregularis*) study occurred from May 18 to October 6, 2020. (A) location of mapped area on the island of Guam. (B) Polygon describing the extent of nontoxic brown treesnake bait placements (240 ha).

Figure 4. Bait tube and bracket emulating a tree branch with a time lapse trail camera set to monitor baits. Monitoring of brown treesnake (*Boiga irregularis*) baits occurred on Andersen Air Force Base, Guam, from May 18 to October 6, 2020.

strate the extent of work conducted to reduce BTS numbers at AAFB. Despite these control efforts, evidence from visual searches and predation of birds tagged with radio-transmitters demonstrates the presence of a population of BTS within AAFB housing (Pollock et al. 2019).

Methods

Bait monitoring

We divided the study area into 32 parcels of approximately even size and assigned them a randomized order. Parcels were only delineated for logistical purposes and were not considered units of replication. For each week of the study, we outfitted 1 or 2 parcels with 12 bait stations temporarily affixed to trees spaced at least 20 m apart. We preferentially selected larger trees known to be used by BTS and såli (ironwood, flame tree, and banyan) and used coconut trees only when other trees were not numerous enough because they are rarely seen being used by BTS on AAFB (S. Goetz, U.S. Geological Survey, and M. Kastner, Iowa State University, personal observation).

We modeled bait stations (Figure 4) on those used by WS for operational BTS control, composed of a 30.5-cm-long × 5.1-cm-diameter plastic pipe with the opening on each end bisected by a 6.35-mm-diameter bolt to preclude larger nontarget animals from entering (Mathies et al. 2011, Clark et al. 2012, Lardner et al. 2013, Clark et al. 2018). These bait stations (i.e., bait tubes) are typically suspended horizontally by each end from existing vegetation or fences with 10–30 cm of nylon cord at approximately chest height (1.3 m above ground level) for ease of servicing.

Because most trees throughout the study area do not have branches at the appropriate height, we fashioned temporary bait tube brackets from 15- and 75-cm lengths of 19 × 38-mm lumber connected perpendicularly by a hinge. For temporary installation on trees, we lashed the shorter length of lumber to the bole of the tree, then pivoted the longer length (arm) on the hinge to where it was level and secured the angle with a length of nylon cord between eye bolts on each length of lumber (Figure 4). We then suspended bait tubes under the arms with plastic clothesline material and mounted an infrared trail camera below the arm on a threaded bolt such that the field of view lined up with the center of the tube. Artificial armatures are commonly used for supporting bait tubes where sufficient vegetation does not occur and for mounting cameras to validate BTS versus nontarget bait takes, and they do not appear to negatively influence bait takes by BTS (Mathies et al. 2011; Siers et al. 2019*b*, 2020*a*).

We used Apeman H68 wide-angle cameras (Apeman International Co., Ltd., Huanan City, Shenzhen, China) powered either by 8 internal AA batteries or an external 12V battery pack (Moultrie Products, Birmingham, Alabama, USA). To minimize the potential for small children and pets to access the baits, we mounted brackets as high as we could while ensuring that technicians could visually inspect the baits (approximately 1.6 m). After we performed trials in all 32 parcels, we repeated the first 9 parcels on the list for a total of 41 trials of 12 baits each (492 bait station locations).

Base housing managers were notified of the study objectives and methods. We were unable to assess the extent to which this information was disseminated to residents.

Bait selection and preparation

Although operational baiting typically uses 5 g AMB, the primary targets of our baiting in AAFB housing are BTS large enough to attack and kill hatchling to adult såli (approximately 10–85 g, respectively). Therefore, we elected to evaluate take rates for 2 sizes of rodent and bird baits including small mouse (13–17 g; SM), large mouse (25–35 g; LM), small bird (10–14 g quail chick; SB), and large bird (25–35 g chicken chick; LB) baits (Figure 5).

All baits contained radio-transmitters. We prepared baits for telemetry by using surgical scissors to cut a small incision in the abdomen of the bait, inserting a very high frequency radio-transmitter into the viscera through the incision and suturing the incision closed with monofilament suture material or fishing line. Transmitters were either 5 g Holohil SB-2 transmitters with external wire antenna trimmed to approximately 10 cm (Holohil Systems Ltd., Carp, Ontario, Canada) or 4 g ATS R1770 transmitters with internal ribbon antenna (Advanced Telemetry Systems, Isanti, Minnesota, USA; Figure 5). All baits for this study were nontoxic (i.e., did not contain acetaminophen).

Field procedures

We randomly assigned the 4 bait types to bait tube locations. All baits contained radio-transmitters and were monitored by cameras. We placed the baits in the center of the horizontal bait tubes with long tongs, and cameras were set to take a photograph every 20 seconds for 24 hours per day because snakes and other small animals do not reliably trigger motion sensors (Urbanek et al. 2019). Baits were checked daily for 4 consecutive days and recorded as: taken; not taken, still in tube; or not taken, but found on ground under the tube. Baits quickly degraded, especially by fly (Diptera) larvae, and by the nineteenth week of the study we had observed that no takes occurred after the third night (consistent with observations of standard AMB; Siers et al. 2019*b*) so we reduced the bait monitoring period to 3 days. If baits were taken, we confirmed the species that took the bait by reviewing photographs from the cameras (Figure 6).

We tracked removed baits via homing on the radio-telemetry signal with handheld receivers and antennae to within as close to the snake's sheltering location as possible. We recorded the global positioning system coordinates of the estimated location, the estimated height from which the signal was coming, and a description of the presumed refugium of the snake (e.g., tree genus). We then attempted to capture the BTS; if unable to secure the snake, we made further attempts over successive days and occasional nighttime visits, recording refugium data each time, until we captured the snake or the transmitter was regurgitated or defecated. If captured, we recorded BTS snout-vent length (SVL), weight, and determined sex by probing for inverted hemipenes (Reed and Tucker 2012).

Figure 5. Variation in brown treesnake (*Boiga irregularis*) baits offered. Clockwise from top left: large bird (25–35 g chicken chick), large mouse (25–35 g), small mouse (10–15 g), and small bird (10–15 g quail chick). Photograph also includes 4-g very high frequency radio-transmitter. This study occurred on Andersen Air Force Base, Guam, USA, from May 18 to October 6, 2020.

Figure 6. Time lapse camera image confirming take of a large bird bait (25–35-g chicken chick) by a brown treesnake (*Boiga irregularis*). This study occurred on Andersen Air Force Base, Guam, USA, from May 18 to October 6, 2020.

Statistical analysis

All statistical analyses were performed in R version 3.5.3 (R Development Core Team 2019). Differences in bait take rates among bait types and sizes were tested with binomial logistic regressions, and pairwise comparisons of these categorical factors were performed with the "emmeans" package.

All work was conducted in accordance with the U.S. Department of Agriculture (USDA), Wildlife Services National Wildlife Research Center Animal Care and Use Committee approved protocol QA-3104.

Results

Bait placement began May 18 and ended October 6, 2020. We placed bait tubes in 492 locations. Ten bait trials were discarded due to human error or equipment malfunction, so we successfully monitored 482 baits (Table 1). Brown treesnakes removed 32 (6.6%) of the baits, but 12 of those were dropped near the bait tube. Thus, BTS removed 20 (4.1%) of the baits from the vicinity of the bait stations with 1 bait appearing to have been regurgitated away from the bait station; in cage trials, all BTS that regurgitated baits with acetaminophen died (Siers et al. 2021), so we assumed

Table 1. Results for 482 nontoxic brown treesnake (*Boiga irregularis)* baits monitored within the residential areas of Andersen Air Force Base, Guam, USA, from May 18 to October 6, 2020, as confirmed by camera monitoring. *N* = number of baits; % = percent of total number of baits monitored.

Bait fate	*N*	%
Taken successfully by snake (confirmed on camera)	19	3.9
Taken by snake but regurgitated (found on ground away from bait tube)	1	0.2
Taken unsuccessfully by snake (confirmed on camera, but bait dropped)	12	2.5
Taken by other species (3 monitor lizards [*Varanus indicus*], 1 coconut crab [*Birgus latro*])	4	0.8
Taken by unknown species (probably cat [*Felis catus*], no camera confirmation)*	1	0.2
Not taken (remained in bait tube)	411	85.2
Not taken (found on ground due to weather or movement by fly [Diptera] larvae)	34	7.0

* A cat had knocked down the bait tube bracket; the bait was gone and may have been taken by the cat, but this could not be confirmed.

Table 2. Summary of 16 brown treesnakes (*Boiga irregularis)* that had taken baits and been successfully tracked to their first refugium. Only 8 of these snakes were successfully captured, so morphometric data are lacking for the remaining 8. SVL = snout-vent length, and height is the estimated height of the snake location above ground. SM = small mouse, LM = large mouse, SB = small bird, and LB = large bird. This study was conducted on Andersen Air Force Base, Guam, USA, from May 18 to October 6, 2020.

SVL (mm)	Mass (g)	Sex	Bait	Height (m)	First refugium
840	79	F	SM	—	Forest outside base fence
899	71	F	SM	3.5	Under *Casuarina* needles on house roof
940	84	M	SM	>2	In *Barringtonia*
958	120	M	SM	1.3	Under needles in crook of *Casuarina*
1,011	105	M	SB	1.8	Hole in *Plumeria*
1,090	188	M	SM	1.5	Coconut palm
1,095	259	F	LM	0	Under shrubbery at base of *Barringtonia*
1,126	297	F	LB	0	Inside decomposing coconut husk
—	—	—	SB	1	Hole in trunk of *Casuarina*
—	—	—	LM	<0	Under *Ficus*, center of tree in leaf litter
—	—	—	LB	<0	Hole under clump of *Ficus* roots
—	—	—	LM	0.3	Hollowed root area of *Casuarina*
—	—	—	SM	2.1	Crevice on side of *Casuarina*
—	—	—	LM	<0	*Casuarina* roots
—	—	—	LB	0.5	*Ficus* roots
—	—	—	SB	0.3	Deep into *Ficus* roots

that 20 BTS would have died if baits contained acetaminophen.

There were only 4 cases of confirmed nontarget take of baits, which included 3 mangrove monitor lizards (*Varanus tsukamotoi*, until recently known as *V. indicus*; Weijola 2020) and 1 coconut crab (*Birgus latro*), for a nontarget bait take rate of 0.8%. One additional bait station was dislodged by a stray or feral domestic cat (*Felis cattus*); the transmitter was found on the ground, but the small mouse bait was missing and may have been ingested by the cat. The remaining 445 baits (92.2%) were not taken; 34 of these had fallen to the ground beneath the bait tubes either due to wind or movement by fly larvae (maggots).

For the 32 baits that were removed from bait stations by BTS (taken successfully, taken unsuccessfully, or taken and regurgitated), we found no statistical differences among take rates for small mouse (n = 10), large mouse (n = 9), small bird (n = 7), and large bird (n = 6) bait types (P for all pairwise comparisons >0.7), between large (n = 15) and small (n = 17) baits (P = 0.733), or between mouse (n = 19) and bird (n =13) baits (P = 0.275). Of the 12 baits dropped, there was no difference among baits in rates at which they were dropped, either by type (bird or mouse; P = 0.251) or by size (small or large; P = 0.570). Power for all of these statistical tests was extremely limited due to the minimal number of bait takes.

Of the 19 baits confirmed to have been successfully taken by BTS, we were only able to recover 8 BTS (Table 2). Sizes ranged from 840–1,126 mm SVL and averaged 995 mm. Their weight (body mass) ranged from 71–295 g, averaging 150 g. The 6 smallest snakes recovered (840–1,090 mm) had taken small baits (all small mice except a 1,011-mm BTS that took a small bird). Suggestively, the largest snake in the data set (1,126-mm female) took a large bird bait, and the next-largest snake took a large mouse; all other recovered snakes had taken a small bait. However, these data are too sparse to draw conclusions about bait preferences by snake size class.

We were able to track 16 snakes to their site of first refugium following bait take (Table 2). Including subsequent tracking efforts, we recorded 65 total locations. One snake entered a gap in an air conditioning vent and moved into the interior portion of the vent in an uninhabited house, where it remained for several days. One snake was tracked to either the roof of a house or a storage closet for 1 night (the transmitter was found defecated in the yard the following day); if the snake was in the storage closet, increased potential existed for accidental inclusion in a household goods shipment. One snake was tracked to a pile of ironwood needles on the roof of an uninhabited house. All other refugium locations were in trees or associated landscaping.

Spatially, BTS bait takes were primarily located near forest edges, with few to no takes in the center of the study area (Figure 7). The 3 nontarget takes by mangrove monitors overlapped with areas of take by BTS, while the only observation of a take by a crab (a small coconut crab) occurred near the center of the study area, far from any BTS bait take activity. The suspected bait take by a cat occurred next to the commissary, far from the residential areas.

Discussion

Prior to BTS suppression activities, dead mouse bait take rates for forested areas on Guam typically range from approximately 50–85% (Savarie et al. 2001; Clark et al. 2012; Siers et al. 2018, 2019*b*, 2020*a*). In areas undergoing sustained BTS control, typically along forest edges, take rates range from approximately 10–25% (Savarie et al. 2001; Clark et al. 2012; Siers et al. 2018, 2019*b*; A. Collins, USDA Wildlife Services, unpublished data). During this study, BTS bait take rates in the AAFB Housing Area and adjacent urban infrastructure were very low at only 4.1%. This may be due to lower density of snakes in urban areas compared to forests (e.g., differences in habitat preference, artificial structures, limited prey availability) and partially influenced by limited BTS management where forest edges border the study area. Additionally, BTS movements may be minimized in the area because urban vegetation is discontinuous and sparsely distributed, making encounters with bait tubes infrequent. Nonetheless, BTS predation in this unnatural habitat continues to reduce såli survival (Pollock et al. 2019).

Visual detection of BTS and såli consumption by BTS are known to occur within the central area that produced low to no bait takes. It may be that snakes newly entering the housing area

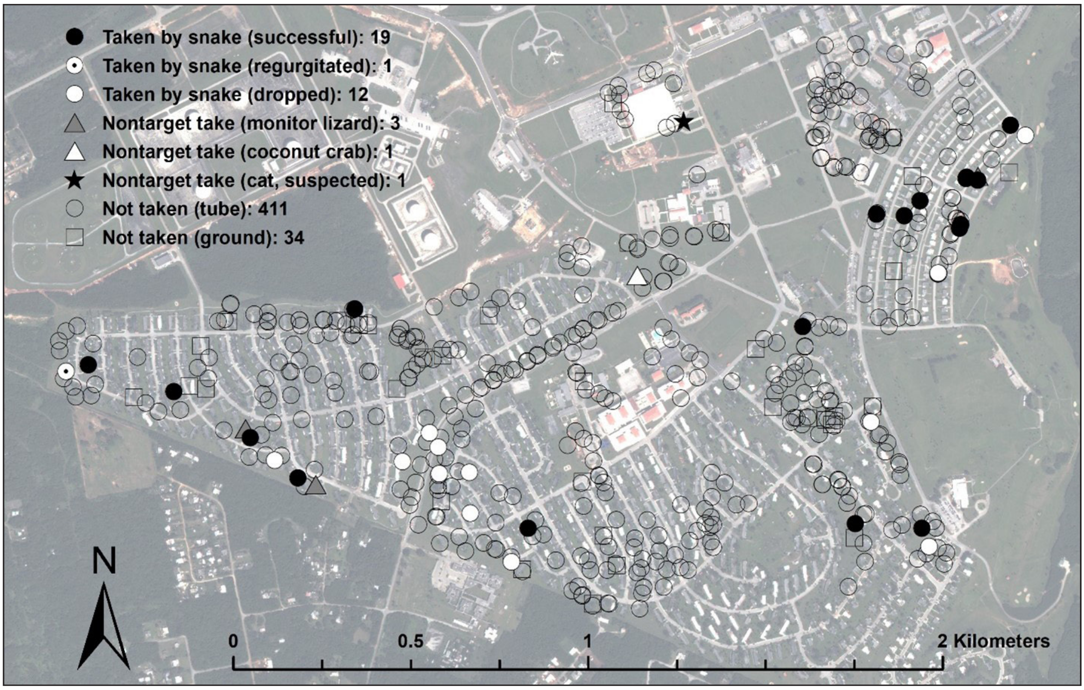

Figure 7. Spatial arrangement of baiting results for 482 nontoxic brown treesnake (*Boiga irregularis*) baits (mice and chicks) placed within and around residential areas on Andersen Air Force Base, Guam, USA, from May 18 to October 6, 2020.

from nearby forest habitat—where live bird and rodent prey are acutely suppressed due to sustained predation by BTS—are more likely to consume carrion baits, as opposed to longer-term resident BTS that have adapted their foraging strategies to preferentially prey on live birds rather than carrion baits.

We observed no strong pattern of BTS preference for bird over mouse baits at the population level; however, the sample size of BTS bait takes was too low to draw any firm conclusions. Even if mouse bait take rates were higher than for bird baits, it would remain unclear whether the individual snakes that took bird baits would have eventually taken a mouse bait. Prior experimental research in a captive environment suggests that BTS that are captured with a bird in their digestive tract are significantly less likely to ingest a mouse bait and that this behavior is repeatable across trials, supporting individual-level preference may exist (Nafus et al. 2021). However, captive behavior is not always a good measurement of behavior in the wild, so this sort of individual heterogeneity remains an important research area. Where protection of birds is a priority, it might be advisable to include birds within a baiting program.

During this study, only 12–24 bait tubes were in the field at any time. Despite the low rate of bait takes, we speculate that the removal of 20 snakes at relatively low effort may still make a meaningful impact over time, especially with increased bait tube applications. During the next phase of this study, proposed plans include staging 50 bait tubes to be continually maintained within each of 4 study plots and baited twice weekly to reduce predation on såli. At 400 baits per week over a 6-month period, 9,500 baits would be set out in the field. At our observed take rate of 3.9%, approximately 374 BTS would be removed, but take would likely decline if BTS density was reduced; valid BTS density estimates for AAFB areas are not available. Data collection during this future phase provides an opportunity for us to further evaluate bait take rates under sustained suppression and preference for bait types.

Nontarget interactions with BTS baits were very low (3 monitor lizards and 1 coconut crab). We observed or recorded no other nontarget interactions with baits, including birds such as såli, except possibly a house cat.

Additional measures may be needed to reduce the number of baits that fall from bait tubes due to wind and invertebrate activity to prevent availability of toxic baits on the ground; however, in all of these cases, the bait was found on the ground under the tube and none were taken by nontargets. One cat was known to have interfered with a bait tube and camera setup, and the bait was not found, so it was assumed that the cat could have possibly eaten it. Johnston et al. (2002) estimated the LD50 of acetaminophen for cats to be 361 mg acetaminophen per kg of cat body mass. A small adult cat weighs approximately 3.5 kg; therefore, a cat would need to consume 1,263 mg (16 tablets) to reach the LD50. Thus, it is believed that even if some free-ranging cats take acetaminophen baits, they will not likely be killed. It is possible that the cat might not have been able to access the bait if the temporary bait tube bracket had not failed to hold the weight of the cat.

Stray and feral cats are sporadically removed from AAFB, but animal control measures were paused at the time of this study due to the COVID-19 pandemic response (M. Hall, U.S. Navy Joint Region Marianas, personal communication). Because this single occurrence was next to the commissary, far from residential areas, it is likely that the cat was not a pet at the time.

We observed no human interactions with bait tubes or camera devices. This was encouraging from a safety standpoint. However, AAFB residents are accustomed to centralized property management, landscaping, and animal control so are possibly less likely to interfere with BTS removal activities than residents in other areas of Guam. The use of baits in off-base urban and residential areas on private lands has yet to be evaluated for domestic animal or human health risks. Any baits that are removed by nontargets or fall from bait stations are quickly consumed by scavengers or detritivores, as are the carcasses of BTS killed by acetaminophen intoxication (Smith et al. 2016). Microbial action rapidly breaks down acetaminophen in soils (Li et al. 2014).

Following intensive aerial broadcast of acetaminophen baits on Guam (120 80-mg tablets per ha over repeated applications; Siers et al. 2020*a, b*), tests showed no acetaminophen residues in aquifers beneath the action areas (USDA Wildlife Services, National Wildlife Research Center, Chemistry Lab Unit unpublished reports 16-040 and 18-033), indicating no evidence of percolation through soils. Therefore, it is unlikely that the use of Acetaminophen for Brown Treesnake Control by certified applicators within the restrictions of the EPA label (as required by law) would pose meaningful risk to human or environmental health.

Acetaminophen toxicity in humans, primarily renal tubular necrosis, hypoglycemic coma, and hepatotoxicity, is common because the medication is so readily available and used widely by >60 million Americans on a weekly basis as a nonprescription analgesic; overdoses are often due to dosing confusion with the variety of formulations (National Institutes of Health [NIH] 2021). Acetaminophen is the most common cause of acute liver failure in western countries (Larson 2007, Chun et al. 2009) and in children taking it long-term in doses of >75 mg/kg day (Heard et al. 2014). Hepatotoxicity typically occurs in children given the wrong calculated dosage, fasting or critically ill patients with concurrent illnesses such as alcoholism and malnutrition, or after suicide attempts with a dosage of >7.5 g (generally >15 g; NIH 2021). Even so, use according to prescribed dosages without compounding illnesses is deemed safe. For humans, the amount used in AMB (80 or 160 mg) is low enough that toxicity is highly unlikely; even in the unlikely case that a tablet were pried from a putrefying bait and taken by a child, it would not have a negative effect.

Potential nontarget animals in urban areas of Guam include primarily animals that would eat an AMB such as those discussed above (dogs, cats, monitor lizards, rats, and coconut crabs; exposure risks evaluated in Johnston et al. 2002), or those exposed environmentally from baits dropping and potentially dissolving in water. Additionally, several bird species may take the mouse baits if available on the ground, but not from the bait tubes. The fact that all baits observed to have fallen from our bait stations were found intact on the ground indicates no appreciable removal of fallen baits by other species. However, military housing on Guam typically has many fewer feral animals than other urban locations on the island. Acetaminophen is not highly toxic to any mammals or birds but is toxic to aquatic invertebrates (Johnston et al. 2002). If an 80-mg tablet were to fall in a gallon of water, it would be equal to about 20 ppm in the water, not enough to be lethal to the most sensitive invertebrate tested. Thus, it is unlikely that acetaminophen would produce a toxic environment for most species, even if a tablet were to drop directly in a pond. Nonetheless, current label restrictions prohibit uses that could enter water sources or aquatic systems without further testing of potential environmental effects.

Many snakes remained in the same location for several days after taking a bait, consistent with previous studies documenting post-feeding dormancy (Siers et al. 2018, Wagner et al. 2018). Although most BTS refugium locations were in trees or other elements of landscaping, 3 snakes were tracked to houses (i.e., roof, air conditioning vent, and potentially a storage closet). Any BTS taking refuge in or near a house demonstrates a potential risk for accidental inclusion in outbound household goods shipments. This information is important for management to develop better control strategies within housing or warehouse areas.

The largest BTS on Guam currently tend to be found in urban habitats (Savidge 1991; Siers et al. 2017*a*, *b*). Unfortunately, the standard 80-mg acetaminophen dose has recently been demonstrated to be much less effective for extremely large BTS, which may require much larger doses to ensure lethal removal with a single bait (Siers et al. 2021). Based on these research results, the EPA recently approved a modification to the acetaminophen label to allow for placing multiple tablets in larger baits when specifically targeting very large snakes. Application of baits with larger doses of acetaminophen would change the risk profile for larger nontargets that might ingest baits (e.g., cats or dogs), which should be carefully considered prior to implementation in residential areas. Reliable removal of extremely large snakes from urban areas might require including alternative methods such as trapping (Siers et al. 2021).

The AAFB housing area is unusually regimented and well-maintained, and not representative of many residential areas on Guam. Conditions in other housing areas are expected to vary much more widely in landscaping, sanitation, human and domestic animal interactions with bait stations, and proximity to BTS prey and habitat resources. Application of similar methods in a range of residential conditions may be advisable if use of acetaminophen baits for BTS control in private or public urban areas is to be considered more widely throughout Guam. The EPA approval for use of the WS-registered acetaminophen tablet for BTS control is currently limited to employees of the U.S. state and federal governments, the Government of Guam or the Commonwealth of the Northern Mariana Islands trained in BTS control, or persons under their direct supervision. Any use of alternative acetaminophen products by other persons as a snake or reptile pesticide would be illegal under the Federal Insecticide, Fungicide, and Rodenticide Act (7 U.S.C. ch. 6 § 136 et seq.).

Management implications

If demonstrated safe and effective, and approved by federal and local laws, use of acetaminophen for BTS control by applicators authorized by the label could be brought "closer to home." Reduction of BTS in urban areas would result in improved quality of life for residents, reduced economic impacts from power outages, improved survival of wildlife occurring in urban environments, and would likely contribute to a reduction in BTS abundance in adjacent forest areas.

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Associate Editor: S. Nicole Frey

Shane R. Siers is a wildlife ecologist and conservation biologist working primarily on invasive vertebrates in island ecosystems, including rodents, mongooses, birds, and reptiles. He has worked on invasive brown treesnake research since 2006 and is currently a supervisory research wildlife biologist with the U.S. Department of Agriculture National Wildlife Research Center. After 6 years of active duty in the U.S. Marine Corps and 10 years in the zoo field, he completed his B.S. degree in biology at the University of Massachusetts Boston, M.S. degree in biology at the University of Missouri-St. Louis, and Ph.D. degree in ecology at Colorado State University. He escapes snakes by going underwater, filming nudibranchs and other small creatures, and posting the videos to his website (www.macronesia.net).

Juan-Carlos Mungaray is a Mexican American hailing from the midwestern state of Iowa. He graduated from Iowa State University with a B.S. degree in biology and environmental science. Since graduating in 2019, he has found himself in the U.S. territory of Guam, having worked on multiple projects around the Marianas. Currently, he is a research associate at the University of Guam working on fruit bats and native plant research with the Center for Island Sustainability. He has a strong interest in conservation problems such as invasive species, especially at the disease-causing level such as chytrid fungus, white-nose syndrome, and avian malaria. He plans to pursue a graduate degree once his time in Guam is over.

Jordan I. S. Barcinas (photo unavailable) is a biological science technician for U.S. Department of Agriculture (USDA) Animal and Plant Health Inspection Service (APHIS) Wildlife Services. He has >2 years of experience in the agency under both USDA APHIS Wildlife Services and as a University of Guam research assistant contracted to the USDA National Wildlife Research Center. He graduated from the University of Guam in 2021 with a bachelor's degree in applied-biology.

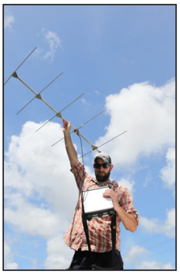
Jeried E. Calaor was born and raised in Guam and obtained both his B.S. and M.S. degrees in biology from the University of Guam. His interests in natural and cultural resource conservation blossomed while working directly with several ESA-listed species, such as the Mariana fruit bat and green sea turtle, during his time working at Guam National Wildlife Refuge – Ritidian Point. He spent several years as a field technician on the Brown Treesnake Research Support Team doing work that he hopes will one day be used to suppress brown treesnakes so that native avifauna of Guam can be reintroduced. When he is not sitting behind his desk, he enjoys spending time outdoors.

Rachel M. Volsteadt was born and raised on Guam and has been working on various brown treesnake projects with the U.S. Department of Agriculture Wildlife Services Guam State Office and National Wildlife Research Center since 2012. In addition to brown treesnake work, she gained a couple years of experience trapping feral swine on Guam Department of Defense lands and private residences. When she is not wrangling brown treesnakes, she enjoys exploring the jungles of Guam, lounging on a beach, looking for the next new country to travel to, and watching all of the latest movies.

Martin Kastner is a Ph.D. student in the Department of Fish and Wildlife Conservation at Virginia Tech. His academic and professional backgrounds are in restoration ecology and endangered species recovery. He is currently studying interactions between invasive brown treesnakes and native birds on Guam, with the ultimate goal of recovering native bird populations. He is dedicated to promoting local community involvement in restoration efforts on the island.

Authors continued on next page...

Scott M. Goetz is currently a biologist with the U.S. Department of Agriculture National Wildlife Research Center. He obtained an M.S. degree from Old Dominion University and a Ph.D. degree from Auburn University. He has conducted research on invasive reptile and amphibian species throughout his career with work primarily focused on bioinvasions in Florida and on the Marianas archipelago in the Pacific. His work takes an applied approach—attempting to better understand the natural history and ecology of invasive species in service of improving methods to contain and minimize the impacts of already established nonnatives. He is interested in refining early detection and rapid response networks, in particular, applying decision science principles to help determine the best use of limited response resources.



Melia G. Nafus (photo unavailable), Ph.D., is a research ecologist with the U.S. Geological Survey Pacific Island Ecosystems Research Center.

Thomas C. Hall earned an M.S. degree in wildlife ecology at Oklahoma State University and has been with the U.S. Department of Agriculture (USDA) Animal and Plant Health Inspection Service (APHIS) Wildlife Services (WS) for 37 years. He started the Guam WS Brown Treesnake Program in April 1993 and has an interest in invasive species and island biogeography. He currently is an environmental protection specialist for USDA APHIS Policy and Program Development and is completing documents related to risk assessments for wildlife damage management methods including acetaminophen baits.