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An effective chemical deterrent for invasive Cuban treefrogs

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Abstract: Introduced vertebrates have a variety of impacts on ecosystems and economies, and many cause problems for humans. One such problem is the loss of electrical power when invasive animals cause short circuits in power-transmission equipment. Cuban treefrogs (*Osteopilus septentrionalis*) are known to cause power outages and are a nuisance to humans when they invade homes and defecate on doors and windows. These large, slightly toxic treefrogs were introduced into Florida from the Caribbean. They now occur throughout the peninsula of Florida and are spreading to other states in the Southeast. We used refuge-choice experiments to test the effectiveness of Sniff 'n' Stop™ animal deterrent to exclude Cuban treefrogs from enclosed spaces, such as utility switchgear boxes. We found that the deterrent was effective and showed potential as a low-cost means to prevent frog-related power outages and reduce conflicts with residents in the urbanized areas preferred by these invasive frogs.

Key Words: Cuban treefrog, deterrent, human–wildlife conflicts, invasive species, nuisance wildlife

Invasive species are plants, animals, and microbes found outside of their native ranges that negatively impact the ecology, economy, or quality of life of humans (National Invasive Species Council 2008). The potential for interactions between humans and a great variety of invasive animals is exacerbated because the urbanization of native habitats enhances the invasion success of introduced wildlife (Lockwood et al. 2007). Well-known examples of invasive animals that exploit human-modified environments include black rats (*Rattus rattus*), European house sparrows (*Passer domesticus*), and red fire ants (*Solenopsis invicta*).

Conflicts between humans and invasive animals are manifested in many ways, including a staggering annual economic impact on businesses and taxpayers in the United States. One estimate of the annual costs associated with losses, damages, and efforts to control invasive species found the cost of 6 invasive mammals to be $37 billion (Pimentel et al. 2005). Invasive animals may directly threaten human health through the spread of disease and envenomation with toxins from bites or stings. For example, feral hogs (*Sus scrofa*) are common agricultural pests that carry numerous diseases transmittable to humans (Clay 2007, Hartin et al. 2007, Kaller et al. 2007). Other invasive animals, such as brown treesnakes (*Boiga irregularis*) and monk parakeets (*Myiopsitta monachus*), indirectly affect humans by causing disruption of electrical service, which results in economic loss to utility companies and businesses (Fritts and Chiszar 1999, Pimentel et al. 2005, Pruett-Jones et al. 2007, Avery et al. 2008).

During the past decade, Cuban treefrogs (*Osteopilus septentrionalis*) have emerged as an invasive species that is responsible for power outages in central Florida. Cuban treefrogs are native to Cuba, Cayman Islands, and Bahama Islands. They have been introduced in Florida, Lesser Antilles, Virgin Islands, Puerto Rico, and Hawaii (Meshaka 2001, Lever 2003). They are notorious hitchhikers, traveling as stowaways in shipping crates, ornamental plants, and vehicles (Meshaka 1996). Cuban treefrogs were first documented in Florida in the 1920s (Barbour 1931), and they dispersed throughout most of southern Florida by the mid-1970s (Meshaka 2001, Meshaka et al. 2004). They are now established throughout peninsular Florida, and continue to expand their range into the southeastern United States (Meshaka 2001, Krysko et al. 2005, Johnson 2007, McGarrity and Johnson 2009).

Cuban treefrogs flourish in human-modified landscapes, such as urban and suburban communities (Meshaka 2001), resulting in human–wildlife conflicts. During the day, they seek shelter in enclosed spaces under shutters and around patio doors; by night, they emerge to feed on insects attracted to lights on homes and other buildings. They defecate on walls and windows, causing unsightly stains (Meshaka 2001; S.A. Johnson, University of Florida, unpublished data). Cuban treefrogs are a nuisance to humans, often invading plumbing systems via vent stacks and seeking refuge in toilets or sink drains (Figure 1). The skin of Cuban treefrogs secretes mucus that is noxious to humans and pets. The mucus can burn the eyes and nose, cause an allergic reaction, and trigger asthma (Meshaka 2001; S. A. Johnson, University of Florida, unpublished data).

Cuban treefrogs impact Florida's economy, at least on a localized scale, when they seek refuge in electrical switchgear boxes. They can cause short-circuits and interruptions in power supplies, increasing maintenance costs for electrical utility companies. In some areas of Florida, such outages occur regularly during spring and fall, at an approximate cost of several thousand dollars per incident; the cost to replace a single piece of equipment damaged in an incident in fall 2007 was about $20,000 (S. Perkins, Lakeland Electric Co., personal communication). Development of effective, broad-spectrum deterrents (effective for use with a wide variety of wildlife species) for use by the utility industry may reduce these outages. One potential deterrent, Sniff'n'Stop™ (IFOAM Specialty Products Corporation, Sanford, Fla.; *http://www.sniffnstop.com*), acts by the time-release of odor molecules (Isophorone; MSDS available at *www.sniffnstop.com*) that many species avoid.

Amphibians have the ability to detect and avoid chemical cues (Wells 2007). Exploitation of this behavior may lead to the eventual development of amphibian deterrents for use in management of pest amphibians (e.g., Hagman and Shine 2008). Cuban treefrogs detect and avoid chemical cues from conspecifics in lab trials (Hoffmann 2007) and avoided Sniff'n'Stop in a pilot study we conducted, suggesting that this deterrent holds potential for use with Cuban treefrogs. Therefore, we conducted laboratory tests to evaluate the effectiveness of Sniff'n'Stop as a deterrent for Cuban treefrogs, with the goal of reducing human–wildlife conflicts caused when these invasive frogs seek shelter in electrical switchgear boxes. We also compared the effectiveness of several formulations of Sniff'n'Stop.

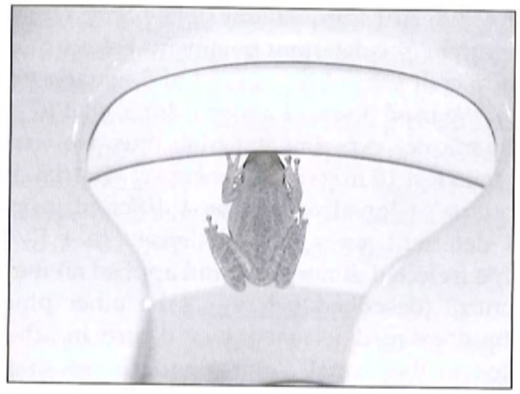

Figure 1. Cuban treefrogs are a nuisance when they invade homes and buildings and hide in toilets, as shown in this photo.

Materials and methods

We used PVC pipe refuges installed at various sites in central Florida to capture 195 Cuban treefrogs. PVC pipe refuges provide a tight, enclosed space that mimics natural refuges preferred by treefrogs, and are commonly used to capture them (Boughton et al. 2000). We placed frogs in holding aquaria filled with 5 cm of moist sand and provided each frog with a vertical PVC pipe refuge (3.8 cm inner diameter, 20 cm long). Each aquarium was covered with a tight-fitting screen lid. We allowed frogs to acclimate to the use of the PVC pipes as refuges for at least 24 hours. We housed similarly-sized frogs together and fed them live crickets (*Gryllus* spp.) *ad libitum* during the acclimation period.

Experimental design

We used standard refuge-choice experiments to evaluate the effectiveness of Sniff'n'Stop at deterring Cuban treefrogs from using enclosed PVC pipe refuges (3.8 cm inner diameter, 20 cm long). We offered frogs a choice between 2 PVC pipes placed vertically at opposite ends of a 40-liter aquarium filled with 5 cm of moist sand. We inserted pipes completely into the substrate so that 15 cm of the PVC refuges extended above the sand. We randomly assigned aquaria to 1 of 4 different formulations of the Sniff'n'Stop deterrent (see deterrent treatments below) or as an overall control, for a total of 5 aquaria per set. We used 3 sets of aquaria, for a total of 15 aquaria per experimental trial; thus, we were able to test 15 frogs simultaneously per trial. In aquaria assigned one of the 4 different forms of deterrent, we applied deterrent to 1 PVC pipe (selected at random), and applied an inert control (described below) to the other pipe. There was no deterrent or inert control in either pipe in the overall control aquaria, so these aquaria served as a check to make sure frogs did not systematically prefer 1 pipe location over the other.

Each refuge-choice trial consisted of 1 period of 24-hours in which frogs were randomly-assigned to each of the 15 aquaria (i.e., 3 sets of aquaria; 1 set = 4 aquaria with deterrents and inert controls plus 1 overall control aquarium). Because Cuban treefrogs are nocturnal, we allowed frogs 24 hours to choose a final refuge site; this enabled them to select a refuge after acclimating to the enclosure during their normal activity period. We recorded their refuge choice (PVC pipe with deterrent, PVC pipe with inert control, another location in aquarium) at the end of the 24-hour period. For the overall control aquaria, frogs were recorded as either being in pipe A or pipe B (neither contained deterrent nor inert control) or in another location. We recorded the sex and length from snout to vent (SVL) for each frog. We conducted 13 replicates of 24-hour refuge-choice trials, for a total of 39 replicates per deterrent treatment (i.e., 13 trials × 3 aquaria per trial for each of the 4 deterrent types plus overall controls), and each frog participated in only 1 trial. Cuban treefrogs are invasive in Florida; therefore, we euthanized frogs immediately after trials by liberal application of 20% benzocaine to each frog's belly. The frogs were then frozen.

Deterrent treatments

We applied Sniff'n'Stop deterrents (4 different formulations) to the inner surface of the PVC refuges as a 2- to 2.5-cm-wide band just above the level of the sand. The foam treatment consisted of a deterrent-impregnated foam strip; we used deterrent-free foam as the inert control. The gel treatment consisted of deterrent microcapsules in a petrolatum matrix; we used plain petroleum jelly as the inert control. The tape treatment consisted of deterrent-impregnated rubber tape; we used 3M Scotch™ Rubber Mastic Tape (#2228), selected for both its similar texture and lack of noticeable odor as the inert control. The epoxy treatment consisted of a 2-part epoxy with repellent microcapsules; we used Loctite® Marine Epoxy (a 2-part epoxy that contains nearly identical ingredients—epoxy resin, isophorone, curing agents) with nondeterrent microcapsules added to duplicate the granular texture, as the inert control. We applied Sniff'n'Stop deterrent treatments and inert controls to PVC refuges only once, just before we started the refuge-choice trials.

Statistical analysis

Data from the overall control aquaria (no deterrent in either PVC pipe refuge) were first tested to verify that frogs used each untreated PVC pipe refuge equally; that is, frogs did not show a preference for either pipe location (west versus east). This was confirmed with a Chi-square goodness-of-fit test. For this test, the observed values were the numbers of frogs that used pipe A (always on the west side of the aquaria) and pipe B (east side of the aquaria). The expected value for the test was the number of frogs expected if pipe-use (A versus B) was completely random (e.g., 50:50 ratio for use of pipes A or B). Thirty-five frogs in the 39 replicates for the overall control aquaria rested in a PVC pipe at the end of the 24-hour trial period; sixteen chose pipe A, and nineteen chose pipe B (4 frogs chose a location outside of the pipes). Therefore, our expected value representing random pipe use was 17.5. Frog use of pipes in the overall control aquaria did not differ significantly from random ($\chi^2 = 0.26$, df = 1, $P > 0.61$).

For the aquaria that received one of the 4 forms of the deterrent, a generalized linear mixed-model was estimated to describe the location of each frog (PVC pipe with repellent versus nonrepellent pipe) after the 24-hour trial period. Generalized linear models differ from the more common general linear models in that the response variable is not assumed to be normally distributed, but can take on a variety of distributions, such as Poisson (for count data), lognormal (for right-skewed data), or in our case, logistic (for binary data). In a generalized linear model, the mean of the response variable is modeled via an appropriate link function, which for binary data is a logit function. As in general linear models, the fixed effects in the model are tested via F-statistics. Our model included a fixed effect for the type of treatment, the covariate SVL and their interaction, and a random effect for trial. We used a type I error level of 0.10 to eliminate the nonsignificant interaction, together with the Quasi-likelihood Information Criterion (QIC; Pan 2001), which is a modified version of the Akaike Information Criterion (AIC) fit statistic that applies to models fit with generalized estimating equations. Whereas the AIC uses the residual variance from the model likelihood, along with a penalty term for each independent variable in the model to measure model fit, the QIC uses the quasi-likelihood function. Infrequently, frogs selected a location in the aquaria outside of the PVC pipes (n = 18; 11% of observations). Because we cannot assume this choice necessarily represented a rejection of the repellent location, we excluded these frogs from further analysis.

Results

Cuban treefrogs selected pipes treated with Sniff'n'Stop only 23% of the time (Table 1). Although none of the forms of Sniff'n'Stop that we tested were 100% effective, they all significantly deterred Cuban treefrogs from using treated refuges. Our final generalized linear mixed-model, which had the lowest QIC (166.5 versus 170.3 for the full model), included only the simple effects of deterrent treatment and SVL. The epoxy treatment was the least effective formulation (31% of frogs in this treatment chose the deterrent pipe), but the effectiveness did not vary significantly among formulations (F = 0.60; df = 3, 36; P = 0.62). Refuge preference was not influenced by frog size (F = 0.01; df = 1,121; P = 0.91).

Discussion

Sniff'n'Stop proved to be an effective deterrent for Cuban treefrogs and, to our knowledge, is the only commercially-available deterrent that has proven effective for use with frogs. All 4 formulations that we tested were effective at preventing Cuban treefrogs from using confined PVC refuges; less than 25% of frogs were resting in deterrent-treated refuges after 24 hours. The success seen in these lab trials suggests that Sniff'n'Stop might be an effective deterrent and warrants field testing. Sniff'n'Stop (U.S. Patent 6,596,204 B1) is commonly used in the field (for other species of vertebrates) without re-application, due to the unique microencapsulation technique used, resulting in the release of the deterrent odor molecules over an extended period of time. However, there is the possibility that Cuban treefrogs could become habituated to the scent and that its effectiveness might wane over time; this must be evaluated. Additionally, potential behavioral effects of this deterrent on native treefrogs should be investigated. Field-testing can be used to evaluate the long-term effectiveness of this product for minimizing human–frog conflicts in a variety of settings.

Although Sniff'n'Stop was not 100% effective

Table 1. Refuge choices of Cuban treefrogs (*Osteopilus septentrionalis*) in experimental trials for each of the 4 formulations of Sniff'n'Stop. Values represent the number of frogs choosing to rest in either the deterrent or nondeterrent pipe or other locations in the aquaria at the end of 24-hour trials for each of the 4 formulations of Sniff'n'Stop. Other locations include sand substrate and aquarium wall.

Deterrent formulation	n (frogs)	Refuge choice: Deterrent-treated pipe	Nondeterrent-treated pipe	Other locations
Foam	39	7	29	3
Gel	39	8	25	6
Tape	39	9	24	6
Epoxy	39	12	24	3
All combined	156	36	102	18

in the laboratory setting, it may be more effective in a closed environment, such as a utility switchgear box. As the odor molecules are released over time in the closed environment of a switchgear box, deterrent levels would likely well exceed the concentrations in our open-ended experimental refuges. Given the low cost of this deterrent (<$15 per unit), it may provide a viable option for prevention of frog-caused power outages.

Lastly, Sniff'n'Stop deterrent holds potential for minimizing conflicts between Cuban treefrogs and humans in urban and suburban settings. Cuban treefrogs thrive in urban settings, where they are able to find plentiful refuges, food, and breeding sites. Sniff'n'Stop may help to exclude Cuban treefrogs from seeking refuge in sheltered spaces on residences, such as vent stacks and spaces behind storm shutters and rain gutters. By restricting the frogs' access to these refuges, Floridians may also be able to reduce the potential for human–frog conflicts.

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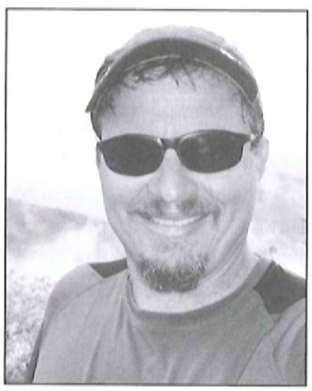

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