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Acetaminophen and zinc phosphide for lethal management of invasive lizards *Ctenosaura similis*

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Abstract Reducing populations of invasive lizards through trapping and shooting is feasible in many cases but effective integrated management relies on a variety of tools, including toxicants. In Florida, using wild-caught non-native black spiny-tailed iguanas *Ctenosaura similis*, we screened acetaminophen and zinc phosphide to determine their suitability for effective population management of this prolific invasive species. Of the animals that received acetaminophen, none died except at the highest test dose, 240 mg per lizard, which is not practical for field use. Zinc phosphide produced 100% mortality at dose levels as little as 25 mg per lizard, equivalent to about 0.5% in bait which is lower than currently used in commercial baits for commensal rodent control. We conclude that zinc phosphide has potential as a useful tool for reducing populations of invasive lizards such as the black spiny-tailed iguana provided target-selective delivery methods are developed [*Current Zoology* 57 (5): 625–629, 2011].

Keywords Acetaminophen, Black spiny-tailed iguana, *Ctenosaura similis*, Florida, Invasive, Zinc phosphide

Florida is home to hundreds of invasive species, including over 30 species of lizard (Meshaka et al., 2004). The black spiny-tailed iguana *Ctenosaura similis* was first released about 30 years ago in southwest Florida on Gasparilla Island, Lee County (Krysko et al., 2003). It now occurs throughout the island, in both Lee and Charlotte counties. The species is most abundant in southwest Florida, but due to additional releases populations are also established in other parts of the state (Townsend et al., 2003a; Meshaka et al., 2004). These iguanas have tremendous reproductive potential, with a single annual clutch containing 12–88 eggs (mean = 43; Fitch and Hackforth-Jones, 1983). One female collected on Key Biscayne, Florida held 82 eggs (Krysko et al., 2003). The black spiny-tailed iguana is popular in the pet trade and is regularly imported to the United States from Central America (<http://www.westcoastiguana.com/>).

Black spiny-tailed iguanas eat landscape plants and enter houses. They also threaten sensitive native flora and fauna. Given its dietary habits and breeding potential, the species presents a genuine threat to native Florida species. Although ctenosaurs are primarily vegetarian, they are opportunists and will eat other lizards,

small birds, rodents, and invertebrates (Fitch and Hackforth-Jones, 1983). Their predatory behavior could potentially affect local populations of imperiled bird species such as the least tern *Sterna antillarum*, snowy plover *Charadrius alexandrinus*, and burrowing owl *Athene cunicularia floridana* (Krysko et al., 2003; Florida Fish and Wildlife Conservation Commission, 2010). These lizards feed on the same native plants as the native gopher tortoise *Gopherus polyphemus*, a threatened species in Florida, and they also occupy gopher tortoise burrows (McKercher, 2001; Krysko et al., 2003; Engeman et al., 2009). There is documented evidence that black spiny-tailed iguanas prey on small gopher tortoises (Avery et al., 2009).

Increasing problems caused by invasive reptiles such as black spiny-tailed iguanas (Townsend et al., 2003b) highlight the need for greater efforts to identify and develop management options. While trapping and shooting have obvious utility, the use of a toxicant can be an important component in the integrated management of invasive species (e.g., Engeman and Vice, 2001). Worldwide, the brown treesnake *Boiga irregularis* is probably the most notorious invasive reptile. Since its

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accidental introduction to Guam about 60 years ago, the brown treesnake has decimated native bird and reptile populations (Savidge, 1987; Rodda and Fritts, 1992) and has major economic impacts as well (Shwiff et al., 2010). Acetaminophen has proven to be an effective oral toxicant for integrated management of the brown treesnake on Guam (Savarie et al., 2001; Shivik et al., 2002). Recently, acetaminophen toxicity was documented in two other reptile species (Mauldin and Savarie, 2010). In this study, we investigated bait acceptance, mortality, and regurgitation in black spiny-tailed iguanas exposed to acetaminophen and to an alternative toxicant, zinc phosphide, in order to assess the potential utility of these compounds in integrated population control programs.

1 Materials and Methods

1.1 Test subjects

United States Department of Agriculture, Wildlife Services (USDA/WS) field personnel on Gasparilla Island, Florida, trapped iguanas and transported them to the USDA/WS/National Wildlife Research Center (NWRC) field station in Gainesville. Each animal was then weighed and placed in an individual plastic cage ($66 \times 43 \times 24 \text{ cm}^3$) with a hinged lid and front viewing window on a commercial 24-cage rack. Inside, at the rear of each cage, we installed a commercially available black plastic shelf. This provided the animals with a dark hiding place below and a convenient basking surface above. A heat lamp mounted outside each cage, approximately 20 cm above the basking shelf, created a temperature range of 25–35°C within the cage, and we mounted an ultraviolet light at the center of each level of the cage rack. Lights, activated by a timer, came on at 10:00h and went off at 20:00h each day.

Maintenance diet, presented in a shallow container, consisted of a variety of chopped vegetables and fruit (e.g. collard greens, red cabbage, broccoli, romaine lettuce, cantaloupe, sweet potatoes, squash, carrots, and apple) augmented daily with 6–7 g of commercial bird of prey diet (Nebraska Brand, North Platte, Nebraska). Each animal received water in a heavy ceramic bowl. We lined each cage with newspaper which was changed daily when the water and food were replaced.

Body mass of field-trapped animals varied, so to provide treatment groups with sets of roughly equal-sized subjects, we ranked each group of iguanas from lightest to heaviest, without regard to sex. Then we employed an online random number generator to assign the animals to treatments (www.randomizer.org/about).

htm). Ctenosaurs were acclimated to captivity for at least 15 days before being used in any trial.

1.2 Toxicant trials: acetaminophen

We presented each test animal with a single dose of 0 mg, 40 mg, 80 mg, 160 mg, or 240 mg acetaminophen. There were eight animals in each test group. For this trial, acetaminophen was prepared in tablet form (40 mg and 80 mg) at the NWRC in Fort Collins, Colorado (Mauldin and Savarie, 2010). To present acetaminophen to the iguanas, we inserted the appropriate dose consisting of 1, 2, or 3 tablets into a red maraschino cherry (approximately 5 g each). Preliminary feeding trials demonstrated that iguanas readily ate the cherries, and the tablets were easily inserted because of the hole created when the cherry pit was removed.

On the test day, we removed the food and newspaper lining from each cage and weighed each iguana. Then we presented each animal with its acetaminophen-filled cherry in a shallow dish. During the “lights on” period, we continuously recorded behavior of two animals with digital surveillance cameras. Greater numbers of animals would have been monitored in this way, but we had just two cameras available. We also observed all animals at half-hour intervals for the first 8 hours of each trial to determine their condition and response to the test substance. Thus, for most animals, we were able to record when bait was ingested and if it was subsequently regurgitated.

Following the test day, each animal resumed its normal maintenance diet. We noted the condition of the animals twice daily for the next 14 days. We did not disturb the test animals during the “lights out” period. Surviving animals were euthanized with carbon dioxide after 14 days.

1.3 Toxicant trials: zinc phosphide

Following the acetaminophen trial, we evaluated zinc phosphide at doses of 0 mg, 6.25 mg, 12.5 mg, 25 mg, 50 mg, 100 mg, and 200 mg. The test material was prepared at the specified dose rates by NWRC personnel in Fort Collins, Colorado by measuring the appropriate amount of zinc phosphide technical powder into gelatin capsules (size #2, Torpac Inc., Fairfield, New Jersey). We presented the test substance inside maraschino cherries and observed the test animals as in the acetaminophen trial. We began with 200 mg and reduced the dose level by half until we recorded no mortality. Because of consistent responses among test animals initially, and at subsequent dose levels, we opted to reduce the number of animals in the trial. Thus, we tested four animals at each level instead of eight.

2 Results

2.1 Acetaminophen

Except for three of eight animals that died in the 240-mg dose group, all iguanas survived the acetaminophen treatments. The animals that died received a mean acetaminophen dose of 709 mg/kg ($SE = 201, n = 3$) compared to a mean of 730 mg/kg ($SE = 187, n = 5$) for the survivors in the 240 mg treatment group (Fig. 1). By day 7 post-dosing, the three animals that subsequently succumbed exhibited marked lethargy and diminished food consumption. Death occurred 9, 11, and 12 days post-treatment, respectively. We observed no adverse behaviors in animals that received sub-lethal doses of acetaminophen. No test animal regurgitated.

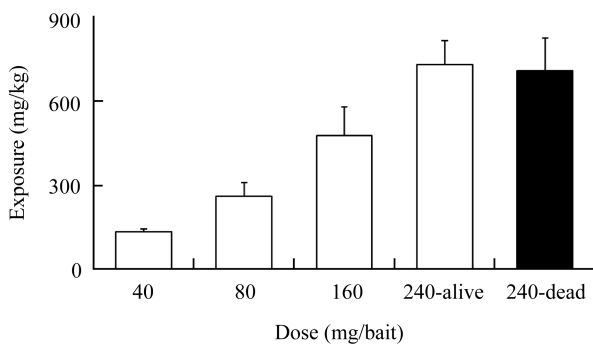


Fig. 1 Singly caged adult black spiny-tailed iguanas each ingested 1 of 4 doses of acetaminophen

Eight animals per treatment group. Three iguanas given 240-mg acetaminophen baits died; all other test animals survived. Capped vertical bars denote 1 *SE*.

2.2 Zinc phosphide

Iguanas received zinc phosphide doses ranging from 13 mg/kg to almost 488 mg/kg (Table 1). With one ex-

ception, all animals exposed to zinc phosphide doses ≥ 25 mg died. The surviving individual in the 50-mg group regurgitated its cherry bait. Examination of that regurgitated cherry and capsule revealed what appeared to be intact zinc phosphide granules.

Time from ingestion of zinc phosphide bait to death ranged from 3.5 h to 174 h, and varied with the dose rate (Kruskall-Wallis H statistic = 9.19, $df = 3, P = 0.027$; Table 1). There was no difference in time to death among the animals that regurgitated and those that did not (Kruskall-Wallis H statistic = 0.01, $df = 1; P = 0.906$). Across all treatment groups, 15 animals regurgitated the bait cherry. Time to regurgitation varied inversely with the dose rate (Kruskall-Wallis H statistic = 9.03, $df = 3, P = 0.029$; Fig. 2).

Other than regurgitation, there was no outward sign of distress or discomfort in ctenosaurs that ingested zinc phosphide. Affected animals remained quiet and generally rested under the shelf at the rear of the cage.

3 Discussion

Ctenosaurs responded very differently to the two test compounds. Zinc phosphide was much more toxic than acetaminophen, and whereas more than half of the animals given zinc phosphide regurgitated, none did in the acetaminophen trials. Time to death was greater in the acetaminophen-treated subjects than for those given zinc phosphide.

Mauldin and Savarie (2010) reported that acetaminophen doses of 522–2438 mg/kg and 263–703 mg/kg were uniformly lethal to juvenile Nile monitors *Varanus niloticus* and Burmese pythons *Python bivittatus*, respectively. We did not obtain such uniformity of response; three ctenosaurs that died ingested acetaminophen

Table 1 Toxicity of zinc phosphide (ZnP) to black spiny-tailed iguanas at various dose rates ($n = 4$ per test group)

ZnP dose (mg/bait)	Body mass (g)		Dose (mg ZnP/ kg body mass)		Time to death (h)			Time to regurgitation (h)		
	Mean	<i>SE</i>	Mean	<i>SE</i>	Mean	<i>SE</i>	<i>n</i>	Mean	<i>SE</i>	<i>n</i>
0	342.5	66.4	0	0	n/a	n/a	0	n/a	n/a	0
6.25	500.8	56.0	12.9	1.3	n/a	n/a	0	31.0	1.7	4
12.5	475.5	35.0	26.7	2.0	n/a	n/a	0	19.8	1.7	4
25	444.8	70.8	60.8	10.0	81.7	20.9	4	4.8	n/a	1
50	445.0	55.2	117.2	13.1	40.4	7.4	3	7.1	1.6	2
100	420.2	17.8	239.5	10.8	74.8	33.3	4	unknown	n/a	1
200	420.0	37.0	487.8	44.4	16.2	6.2	4	3.8	0.9	3

Animals were caged individually and offered a single capsule of toxicant inside a maraschino cherry.

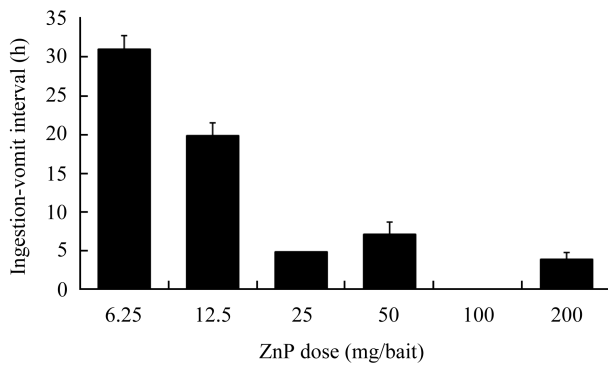


Fig. 2 Singly caged adult black spiny-tailed iguanas each ingested 1 of 6 doses of zinc phosphide

Four animals per treatment group. Most animals regurgitated the bait. Time from bait ingestion to regurgitation was inversely related to the zinc phosphide dose level. Capped vertical bars denote 1 SE.

doses that averaged 709 mg/kg while five surviving animals averaged 730 mg/kg. We are not sure why the ctenosaurs appeared to be less sensitive to acetaminophen than the monitors or pythons. Body size might be a factor as the adult ctenosaurs in our trials were 3–4 times larger than the juvenile pythons and monitors used by Mauldin and Savarie (2010). Another factor could relate to the diet of the respective species. Ctenosaurs are primarily herbivorous whereas pythons and monitors are carnivores. Regardless, based on our findings, the amount of acetaminophen needed to kill a normal adult ctenosaur exceeds practical limits for field applications.

The lowest toxic zinc phosphide dose we administered to a single iguana was approximately 40 mg/kg body mass. Except for some species of bird, published median acute oral toxicity values for other species generally are <50 mg/kg (Casteel and Bailey, 1986; Johnson and Fagerstone, 1994). Various studies have demonstrated that zinc phosphide has low secondary toxicity, so there appears to be little risk to scavengers and predators that might feed on ctenosaurs killed with zinc phosphide (Schitoskey, 1975; Tietjen, 1976¹; Johnson and Fagerstone, 1994).

With one exception, all iguanas that regurgitated after ingesting zinc phosphide bait ≥ 25 mg died. Emesis results from irritation of mucous membranes by phosphine gas which is generated as zinc phosphide is hydrolyzed by stomach acid (Tiway et al., 2005). Because not all animals have the ability to regurgitate and zinc phosphide is very toxic to most forms of animal life, regurgitation cannot be relied on to provide nontarget

species safety from direct primary poisoning (Poppenga et al., 2005). Additionally, regurgitated bait potentially could retain active zinc phosphide that if ingested would pose a danger to nontargets. Because the time to regurgitation was inversely related to dose rate, using the lowest possible effective dose (25 mg) will maximize the time the toxicant is inside the target animal and minimize the likelihood that active material will be contained in the regurgitant.

To use zinc phosphide safely and effectively, the appropriate delivery system must be in place. In the field, a sweet red bait, such as the maraschino cherry that we used in cage trials, will likely attract not only ctenosaurs but various non-target species such as birds and gopher tortoises. We recommend presenting the bait in bait stations that will exclude non-target species. A 25-mg zinc phosphide capsule within a 5-g maraschino cherry constitutes a 0.5% (active ingredient) bait. Most zinc phosphide-based rodent baits are 2% formulations (e.g., ZP® Rodent Bait, Bell Laboratories, Inc., Madison, Wisconsin; USEPA Reg. No. 12455-18). Thus, for control of black spiny-tailed iguanas, the application of zinc phosphide would likely be lower than is currently used for routine commensal rodent control. Provided it can be delivered safely and selectively, zinc phosphide is a potentially useful tool for reducing populations of invasive spiny-tailed iguanas and possibly other invasive reptiles.

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