Herpetological Review

Using a Wildlife Detector Dog for Locating Eastern Indigo Snakes (*Drymarchon couperi*)

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TECHNIQUES

Herpetological Review, 2010, 41(4), 437–442.

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The Eastern Indigo Snake (*Drymarchon couperi*), a large (adults from 1.5–2.6 m total length [Stevenson et al. 2009]) diurnal species, is imperiled and federally listed as "Threatened" due to population declines attributable to habitat loss/fragmentation and declining Gopher Tortoise (*Gopherus polyphemus*) populations (United State Fish and Wildlife Service [USFWS] 2008). In southern Georgia and portions of its Florida range, *D. couperi* is closely associated with xeric sandhill habitats and Gopher Tortoise burrows, and adults often use tortoise burrows for overwintering sites (Diemer and Speake 1983; Hyslop et al. 2009a; Stevenson et al. 2003). The burrows of adult tortoises average 4.5 m long and 2 m deep (Diemer 1992), but in aeolian sand ridge habitats (see Ivester and Leigh 2003) adjacent to blackwater streams in southeastern Georgia, burrows are commonly 6.1–9.1 m in length (D. Stevenson, unpubl. data). Adult *D. couperi* are frequently surface-active during the winter and may bask or shed their skins near burrows, or move between burrows, during periods of mild (10.0–16.7°C) temperatures (Speake et al. 1978; Stevenson et al. 2009). Although habitat use is varied and less associated with sandy habitats in peninsular Florida, *D. couperi* occur in xeric uplands and regularly use tortoise burrows as far south as south central Florida (Layne and Steiner 1996). Because of the extensive time they spend in tortoise burrows and other below-ground refugia, adult *D. couperi* are difficult to locate during field surveys. Developing reliable survey methods for this species is an important priority for *D. couperi* research and monitoring efforts (USFWS 2008).

Recent studies demonstrate that detector dogs have potential as a non-invasive method for locating target wildlife species in their natural habitats (Nussear et al. 2008; Reindl-Thompson et al. 2006; Smith et al. 2003). However, few studies have used detector dogs to survey for snakes. Klauber (1956) mentions a "hound" from Florida that was trained to trail rattlesnakes (*Crotalus adamanteus*) and "bay" them when found; this dog located ca. 500 rattlesnakes in two years. Detector dogs located Brown Tree Snakes (*Boiga irregularis*) in cargo (Engemann et al. 1998, 2002), and, recently, the New Jersey Division of Fish and Game, Endangered Species Program used a trained dog to locate Northern Pine Snakes (*Pituophis m. melanoleucus*) adults and eggs (Dave Golden, pers. comm., 2007).

In an effort to develop an efficient and accurate survey method for locating *D. couperi* in the wild, we conducted a pilot study to test the effectiveness of a trained detector dog at locating Eastern Indigo Snakes. We conducted: 1) experimental field trials to evaluate a trained detector dog's ability to find live *D. couperi* and shed skins of *D. couperi* in the species' natural habitat; 2) actual field surveys, using a trained dog, to survey sites known to support *D. couperi*.

Methods

Dog Selection and Training.—We trained a dog (male, Labrador-mix, 5 years old, 30 kg) on loan from PackLeader Conservation Detector Dogs, Gig Harbor, Washington, USA 98329 to locate *D. couperi* using a combination of detection training techniques. We first introduced the dog to the odor of *D. couperi* at PackLeader in Washington State in October 2008. We randomly placed sections of shed skins (from multiple *D. couperi* collected from the wild in southern Georgia and central Florida) in a large field and allowed them to sit for up to 0.5 h. We then allowed the dog to roam the field, and when the dog showed interest in a shed skin the dog was commanded to sit and was rewarded with its play object (tennis ball). We continued similar shed-skin exercises with the dog over the next two weeks until the dog reliably located the samples.

Next, we trained a handler on the fundamentals of working the dog in the natural habitat of *D. couperi*. We conducted this training at the Orianne Indigo Snake Preserve, Telfair County, Georgia on 17 dates in November, 2008. Well-drained xeric sandhills on-site support resident populations of Gopher Tortoises and *D. couperi*. During this phase of training, we continued field exercises with shed skins and also introduced the dog to the scent of live *D. couperi*.

To train the handler, we set up area-search exercises wherein we hid varying numbers (7–14) of *D. couperi* shed skin sections. The trainer accompanied the handler to point out the dog's working style and to explain the effects of environmental factors on scent. In the first series of exercises, the handler and observer knew the location of the hidden shed skins. Gradually, sheds at undisclosed locations were added to the search area, and the size of the search area was increased (to add time and distance to the search).

When the handler had learned the various changes of behavior exhibited by the dog and could accurately determine when the dog was appropriately "indicating" the location of shed skin from a *D. couperi*, shed skins from several other snake species that are sympatric with *D. couperi* (Eastern Coachwhip [*Coluber f. flagellum*], Florida Pine Snake [*Pituophis melanoleucus mugitus*], and Eastern Diamondback Rattlesnake [*C. adamanteus*]) were added to the exercises. When the dog showed interest in the non-target odors (other than rattlesnake), the handler verbally corrected him off the non-target odor and reinforced on the target odor. When the dog showed interest in rattlesnake odor, the handler administered a physical correction and reinforced on the target odor. The dog quickly learned to avoid rattlesnake odor and to ignore non-target species odors. The dog's avoidance behavior (a "sideways look" at the rattlesnake [or rattlesnake shed] followed by a movement away from the area prior to returning to search mode) was clearly distinctive from the change of behavior it exhibited when expecting a reward for finding the target species. Upon locating the target (i.e., *D. couperi* shed skin), the dog "indicated" by expressing a suite of behaviors including sitting and/or remaining stationary, vigorous tail-wagging, and crouching. We continued training in this fashion until the handler was capable of working exercises independently. The handler and dog found more than 90% of the hidden shed skins during these training exercises.

We conducted several exercises near the end of the training period to introduce the dog to the odor of live *D. couperi* and to the holding cages that we used in the Phase 1 field trials (*see below*). As part of this training, we used both empty cages that never held snakes and occupied cages which held live *D. couperi*. We conducted this training so that the handler would have confidence in the dog's ability to separate these odors. In successful exercises, the dog positively "indicated" on the live *D. couperi* as he did above for shed skins of the species.

Phase 1 Trials.—To assess the dog's ability to recognize *D. couperi* scent, we conducted controlled field tests at one site each in Georgia (Joseph W. Jones Ecological Research Center [JERC], Baker Co., Georgia, USA) and in Florida (Apalachicola Bluffs and Ravines Preserve [ABR], Liberty Co., Florida, USA). To control for the possible effects of wild *D. couperi* present on-site, we selected sites not inhabited by *D. couperi* (Gunzberger and Aresco 2007; Smith et al. 2006; D. Printiss, pers. comm., 2008).

We conducted a total of 108 Phase 1 trials (52 at JERC on 23–25 November 2008; 56 at ABR on 3–5 December 2008). At each site, we evenly distributed trials among a total of four treatments for live *D. couperi* and *D. couperi* shed skins, as follows: 1) Above-ground: A live caged snake was placed on the ground surface within 10 meters of a tortoise burrow; 2) Above-ground: A shed skin (free, not caged) was placed on the ground surface within 10 meters of a tortoise burrow; 3) Below-ground: A live caged snake was placed flush with the entrance of a tortoise burrow, or situated a short distance (≤ 0.5 m or less) inside the burrow tunnel; 4) Below-ground: A shed skin (free, not caged) was placed flush with the entrance of a tortoise burrow, or a situated a short distance (≤ 0.5 m or less) inside the burrow tunnel.

For live snake trials, we used adult *D. couperi* (Total length: 140–200 cm) that we captured by hand from sites in Georgia (N = 3 ♂, 1 ♀) or in Florida (N = 2 ♂, 1 ♀). At our Georgia study site (JERC), we used snakes found in Georgia; similarly, we conducted trials at our Florida study site (ABR) using snakes found in Florida. For shed skin trials, we used recent (< 3 months old) shed skins from adult *D. couperi* found in the wild.

When conducting live-snake trials, we placed one *D. couperi* in a specially-designed, escape-proof holding cage (Herpetological Associates, Inc., Dunnellon, Florida, USA). We constructed two different sizes of holding cages so they would fit into tortoise burrows of varying widths. The smaller cages we made measured 18.3 cm wide, 11.2 cm high, by 61.5 cm in length, whereas the larger cage measured 21.0 cm, 13.0 cm, by 101.5 cm in length. We built holding cages with 15 cm plywood ends, and on one end of the cage we installed a small locking door. We made the walls of the cage of rubber-coated hardware cloth with 1.8 cm square openings. So they would fit into tortoise burrows, we constructed the cages so that the top was arched and the bottom was flat, thus approximating the shape of a tortoise burrow in cross-section.

We placed the cage holding the live snake (or the *D. couperi* shed skin) in the selected test location (i.e., on the ground within 10 m of a tortoise burrow, or just inside the entrance of a tortoise burrow). On field trial dates, we dedicated one individual of our study team to the handling and placement of cages holding live snakes and shed skins to minimize transfer of scent odors; forceps were used to position and retrieve shed skins between trials. We concealed our live-snake-in-cage sets by wrapping camouflage netting around the exposed portions of the cage. On above-ground trials, we positioned cages holding live snakes and shed skins so that they were partially or mostly concealed by ground cover vegetation and not readily visible to the dog survey team/handler.

On each individual trial, we had the dog handler, the handler's field assistant, and the dog visit three tortoise burrows: 1) one burrow with a hidden target (i.e., either a live snake above-ground, a live snake below-ground, a shed skin above-ground, or a shed skin below-ground); 2) one burrow that was empty (i.e., no hidden targets), and; 3) one burrow that was either empty (all shed skin trials) or, for all live snake trials, one burrow with a control (i.e., an empty cage hidden above or below-ground—to verify that the dog was not indicating on the cages). We conducted trials in sets of four, in varying order: snake above-ground; snake below-ground; shed skin above-ground; shed skin below-ground. To ensure that the dog was not following human scent or keying on flags, we marked all tortoise burrows used in these field trials with similar-colored flagging tape tied to nearby vegetation, and we had a supporting biologist introduce human scent at all burrows (by rubbing his hand over the sand inside the burrow and on the apron) while setting up trials. We removed the dog and handler from the immediate area (i.e., minimum 100 m distant) when preparing trial sets. We allowed our live snake and shed skin sets to sit for ca. 10–20 minutes to allow some airborne scent dispersal before bringing the dog and handler to the area.

Next, we had the handler lead the dog to each of the three burrows that constituted an individual trial. We classified a particular trial as successful if the dog correctly indicated the presence of an indigo snake or shed; those trials where the dog did not indicate at/near burrows where we had hidden a snake or shed skin we classified as errors of omission; trials where the dog indicated at/near burrows where we had not hidden a snake or shed skin we classified as commission errors. On all Phase 1 trials, the dog handler was accompanied by a field assistant who helped orient her and lead the dog to trial burrow locations.

We used data from these trials to calculate the proportion of trials where the dog successfully signaled on live *D. couperi* or shed skins. We used a Chi-square goodness of fit test to compare dog success among the four treatments (Above Ground-Live Snake, Above Ground-Shed Skin, Below Ground-Live Snake, Below Ground-Shed Skin). We further classified unsuccessful trials by calculating the proportions that were omission versus commission errors.

Phase 2 Trials.—For the second part of our study, the dog and handler conducted 1-h long field surveys for *D. couperi*. We surveyed non-overlapping xeric sandhill sites that supported numerous active/inactive tortoise burrows and resident, overwintering *D. couperi*. We conducted a total of 26 1–h long trials (2–4 trials per survey date) on nine dates from 8–23 January 2009 at a total of seven sites: Fort Stewart Military Installation (FSMI), Bryan Co., Georgia (N = 3); FSMI, Evans Co., Georgia (N = 3); Broxton Rocks Preserve, Coffee Co., Georgia (N = 5); General Coffee State Park, Coffee Co., Georgia (N = 3); Orianne Indigo Snake Preserve, Telfair Co., Georgia (N = 3); Withlacoochee State Forest, Citrus/Hernando counties, Florida (N = 7); Chassahowitzka Wildlife Management Area, Hernando Co., Florida (N = 2).

Prior to beginning the surveys, we presented the dog survey team (comprised of the dog, the dog handler and her field assistant) with an aerial photograph of the survey site, oriented them with respect to nearby primitive roads, wetlands, and other landmarks, and defined the area of potential habitat to be surveyed. We did not flag tortoise burrows. We then directed the dog survey team to search for one full hour; the dog team made a single pass through each survey area, attempting to visit and search all tortoise burrows they could locate. The field assistant used a compass and aerial photo to orient the dog and the dog handler and keep them on a steady compass bearing. The dog team began the survey at the downwind end of the survey area and progressed upwind, maximizing the dog's exposure to possible *D. couperi* scent. We conducted these surveys from mid-morning through mid-late afternoon on clear or rain-free days (i.e., weather conditions that would prove suitable for *D. couperi* surface activity).

If the dog indicated at the entrance of an individual tortoise burrow (suggesting the presence of a live *D. couperi* or recent shed skin within the burrow), we immediately scoped the burrow with a remote video camera attached to a 9 m section of tubing (Gopher Tortoise Burrow Camera, Southern Ecosystems Research, Auburn, Alabama, USA) in an effort to determine the presence of a live *D. couperi* or shed skin. If *D. couperi* were not documented, we placed a large single-opening funnel trap at the mouth of the burrow in an effort to capture any resident *D. couperi* as they exited the burrow. We shaded funnel traps and checked them 2–3 times per day during daylight hours.

Results

Phase 1 Trials.—The detector dog was correct on 91% (98 of 108) of the Phase 1 trials. Overall, the dog was more successful during shed skin trials than during live snake below-ground trials ($\chi^2 = 13.928$, Df = 3, P = 0.003). The dog was correct in all (100%) of 54 shed skin trials—both above-ground and below-ground. Of the live snake trials, the dog was correct 81% of the time (44 of 54 trials), with 88% success (23 of 26 trials) on above-ground trials,

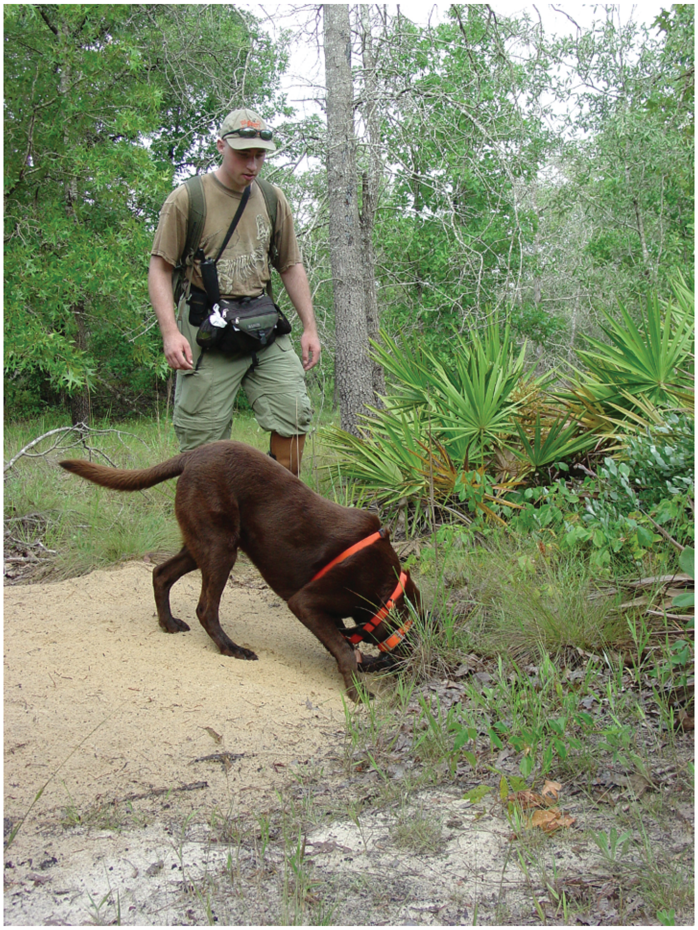

FIG. 1. A specially trained wildlife detector dog ("C.J.") surveys a Gopher Tortoise (*Gopherus polyphemus*) burrow for Eastern Indigo Snakes (*Drymarchon couperi*), Wheeler County, Georgia, USA. Photo by Dirk J. Stevenson.

and 75% success (21 of 28 trials) on below-ground trials. Thirty percent (3 of 10) of the unsuccessful trials were commission errors, all of which occurred on below-ground trials, while 70% (7 of 10) of the unsuccessful trials were omission errors, four of these were below-ground trials.

Phase 2 Trials.—On 26 1-h long Phase 2 trials, the dog team surveyed a total of 496 active/inactive Gopher Tortoise burrows at seven sites. During these surveys, the dog located 11 individual *D. couperi* shed skins and indicated at another 18 tortoise burrows. Seventeen of these 18 burrows were examined with the tortoise burrow camera; *D. couperi* were observed in three separate burrows at distances of 3.7, 6.7, and 7.0 m. All of the remaining burrows were trapped from 2–10 days; no *D. couperi*, or any other snakes, were captured by these efforts.

Discussion

Our study suggests that wildlife detector dogs have value as a field survey method for the Eastern Indigo Snake (*D. couperi*). During controlled field tests (Phase 1 Trials), the detector dog used in this study successful located 81% of live *D. couperi* and 100% of *D. couperi* shed skins.

The dog had very little field training with live *D. couperi* prior to Phase 1 trials, and we strongly suspect that the dog would have performed better with additional training with live snakes prior to these trials. During informal training exercises and field surveys conducted 10 December 2008–27 January 2009 at the Orianne Indigo Snake Preserve (Georgia), the dog found seven individual *D. couperi* (a total of 11 times) on the surface near tortoise burrows, and four *D. couperi* below-ground in tortoise burrows. Additionally, the dog indicated at the entrances of 12 tortoise burrows, four of which were confirmed (by scoping with the tortoise burrow camera) to contain *D. couperi*.

On Phase 2 Trials, the dog confirmed the presence of *D. couperi* at 6/7 sites surveyed, finding three *D. couperi* below-ground inside tortoise burrows and 11 *D. couperi* shed skins above-ground. However, the dog may have falsely indicated snake presence at some of those Phase 2 tortoise burrows where he signaled the presence of a snake below-ground inside the burrow (N = 15). Despite our lack of success in documenting *D. couperi* via burrow camera and trapping surveys, we cannot say conclusively that *D. couperi* were not present in these burrows. Although *D. couperi* are occasionally observed by tortoise burrow camera surveys, scoping burrows is unreliable at detecting the presence of *D. couperi* because the terminus of many burrows cannot be reached with a burrow camera due to burrow length, burrow curvature, inanimate obstacles (e.g., plugs of pine straw, tree roots), or the presence of the resident tortoise partway down the tunnel shaft blocking progress of the camera (Smith and Dyer 2003; Stevenson et al. 2003).

Eastern Indigo Snakes have been documented by the following field methods: 1) visual encounter surveys at or near tortoise burrows in sandhill habitats (Diemer and Speake 1983; Stevenson et al. 2009); 2) single-opening funnel traps placed at the entrances to tortoise burrows (Lips 1991); 3) motion-activated cameras placed at tortoise burrows (Alexy et al. 2003); 4) remote video cameras ("tortoise burrow cameras") to examine the interiors of tortoise burrows (Hipes and Jackson 1996; Stevenson et al. 2003); and 5) drift fence arrays using large "box" traps (Hyslop et al. 2009b). Except for visual encounter surveys, most of the survey methods listed above are either not particularly effective and/or are extremely labor-and-time-intensive (Hyslop et al. 2009b; Smith and Dyer 2003). In southern Georgia, visual encounter surveys at tortoise burrows conducted by experienced herpetologists are often effective in locating *D. couperi* (and *D. couperi* sheds) during the cooler seasons (Hyslop et al. 2009b; Stevenson et al. 2003, 2009). However, human searchers vary in the speed at which they visit burrows, their ability to accurately discern snake tracks, and their ability to spot snakes on the surface (Hyslop et al. 2009b).

Both canine surveys and visual encounter surveys may locate live *D. couperi* or shed skins on the ground near tortoise burrows. During field training, the dog found several live *D. couperi* under natural conditions on the surface that were basking cryptically (e.g., under branches or vegetation) near tortoise burrows. And, in both training and during trials the dog often located small fragments of old *D. couperi* shed skins (hidden under debris or vegetation) that went unnoticed by human surveyors. Our study also indicates that a trained detector dog may locate *D. couperi* deep inside tortoise burrows via olfaction. Our dog was able to survey for snakes ca. 4 hours/day; frequent hydration and rest breaks were needed, and the dog did not perform well in hot weather (i.e., > 23°C). Excessive panting during hot weather affects olfactory abilities and may lower detection rates (Smith et al. 2003). In some disturbed sandhill landscapes where native prickly pear cactus (*Opuntia* sp.) and blackberries (*Rubus* sp.) were especially abundant, the dog experienced difficulties due to abrasions to his paws from thorns. Experienced human searchers (i.e., visual encounter surveyors) often discern indigo snake tracks in the sand of tortoise burrow aprons (Stevenson et al. 2009), and visit and survey tortoise burrows at ca. twice the rate of a dog escorted by its handler and one field assistant (this study).

Possible explanations for dog errors in the field include insufficient training, mistakes due to handler errors (e.g., inadequate search), fatigue, and distractions due to encountering novel scents or wildlife species. Inappropriate weather (e.g., windless days and days following heavy rains) may also have influenced the dog's success.

Detector dog surveys were 61% and 64% successful in detecting the presence of Brown Tree Snakes planted in outbound cargo during 1998 and 1999, respectively (Engemann et al. 2002). In cases where the snakes were not located by dogs, twice as many were missed because the dog did not change its behavior in response to the snake rather than because the handler did not conduct an adequate search pattern (Engemann et al. 2002).

This study, a pilot effort, suggests that specially trained wildlife detector dogs are sometimes able to locate *D. couperi* in the wild, and the effectiveness of these "canine surveys" may be enhanced if conducted in concert with other techniques (e.g., visual encounter surveys of Gopher Tortoise burrows). Additional study will reveal whether canine surveys have value at sites where *D. couperi* is present in very low numbers, during periods when the species is not surface-active, or in habitats lacking Gopher Tortoise burrows. The detector dog used in this study located (under natural conditions) live *D. couperi* on the surface and below ground in Gopher Tortoise burrows, and frequently found shed skins that were overlooked by human searchers. Improved survey methods will enable researchers to better determine the distribution of this imperiled species, especially in regions where the species is now seemingly extremely rare or locally distributed (Florida panhandle [Gunzberger and Aresco 2007]), and may assist in developing a defensible presence/absence survey method for development projects.

Acknowledgments.—We are grateful to the following for their assistance with this project: J. Bauder, B. Callaghan, L. Carlile, T. Crites, B. Davenport, M. Elliott, K.Enge, H. Hall, J. Jensen, J. Macey, P. Moler, V. Morris, S. Osborn, D. Printiss, A. Safer, L. Smith, F. Snow, N. Spencer, and B. Willis-Stevenson. Procedures used in this study followed SSAR "Guidelines for Use of Live Amphibians and Reptiles in Field and Laboratory Research." This study was conducted under scientific collection permits issued to Christopher L. Jenkins from the Georgia Department of Natural Resources (Permit No. 21920) and the Florida Fish and Wildlife Conservation Commission (Permit No. WX08638).

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