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ASSESSMENT OF SUBCUTANEOUSLY IMPLANTED REFLECTOR TAGS FOR RELOCATING MOLE SALAMANDERS (*AMBYSTOMA TALPOIDEUM*)

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ABSTRACT

The highly fossorial habits of adult mole salamanders (*Ambystoma talpoideum*) have made them difficult to study. We assessed the usefulness of subcutaneously implanted reflector tags for relocating mole salamanders in the terrestrial environment using harmonic radar detection. We determined maximum detection depth of tags with varying antenna lengths. In addition, we subcutaneously implanted tags into mole salamanders to assess retention and effect of tags on body weight during a 60-day period. We were able to detect tags of all antenna lengths as deep as 30 cm below the soil. Only 29% of mole salamanders implanted with tags retained them for the entire 60 days. We believe that retention rate of subcutaneously implanted tags needs to be improved before this technique can be widely applied in the field.

Key words: *Ambystoma talpoideum*, Ambystomatidae, harmonic radar detection, mole salamanders, terrestrial habitat, reflector tag, tracking

INTRODUCTION

Mole salamanders are a highly fossorial species, spending most of their life in burrows 3-4 cm below the soil surface (1). This aspect of their life history has made them difficult to study. Little surface activity occurs during the nonbreeding season (1), rendering traditional monitoring techniques such as drift fences and pitfall traps inadequate for assessing land management impacts on this species. Because of their small size, mole salamanders cannot be radio-tracked like larger ambystomatid species such as tiger salamanders (*A. tigrinum*; 2) and spotted salamanders (*A. maculatum*; 3). Radioisotope tags have been used to track a variety of ambystomatid species (1,4) but are expensive and difficult to acquire (5). Harmonic radar detection may provide an effective method to track mole salamanders as well as other small terrestrial vertebrates.

Harmonic radar detection was first developed to locate avalanche victims. The technique consists of a hand held transceiver that emits a 1.7-W continuous microwave frequency of 917 MHz. When the microwaves strike a reflector tag they are reflected at double the frequency (1,834 MHz), which is then detected by the transceiver. Harmonic radar detection has been used to track movements of reptile and amphibian species, including broad-headed snakes (*Hoplocephalus bungaroides*; 6), sharp-tailed snakes (*Contia tenuis*; 7), juvenile spotted salamanders (8), and juvenile box turtles (*Terrapene carolina*; L. Brisbin pers. comm.). The objectives of our study were to: 1) determine the detection range of reflector tags having varying antenna lengths; and 2) to test effects of internal implantation on body weight of mole salamanders and retention rate of reflector tags.

MATERIALS AND METHODS

Our harmonic radar transceiver was a RECCO®8 Avalanche Rescue System (RECCO, Lidingo, Sweden). We constructed reflector tags from Vishay-Telefunken BAT85S Schottky barrier diodes (Newark In One, Chicago, IL) and 37-gauge copper wire. We soldered a wire to each lead as close as possible to the diode and then removed leads with wire cutters (Figure 1). We cut antennae to desired lengths and used a Dremel® rotary tool (Bosch Tool Corporation, Stuttgart, Germany) to remove excess solder material. We coated tags with Plasti-Dip® (Plasti-Dip International, Blaine, MN), a presumably inert compound, to provide further protection from rough edges.

In our first experiment, we tested detection range of tags with antenna lengths of 13, 11, 9, and 7 mm ($n = 1$ each). We inserted tags into 5.2 by 1.8-cm canned sausages to simulate effects of a salamander body cavity and to provide a standard for measuring detection distances of different tag antenna lengths. We placed sausages into small, clear plastic bags to prevent damage when buried underground. We then measured detection distance at depths of 5, 10, 20, and 30 cm. We also determined detection distance on the soil surface, and beneath large (29 cm diameter) and small (13 cm diameter) CWD. We recorded the maximum vertical distance that tags could be read with the transceiver.

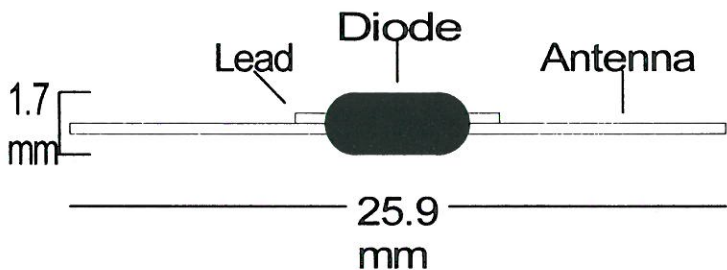


Figure 1. Diagram of subcutaneously implanted reflector tag constructed from Vishay-Telefunken BAT85S Schottky barrier diodes and 37-gauge copper wire implanted in mole salamanders (*Ambystoma talpoideum*).

To test effects of internal implantation and retention rate of tags on mole salamanders, we performed a second experiment. We collected mole salamanders near wetlands at the U.S. Department of Energy's Savannah River Site (SRS), a 78,000-ha National Environmental Research Park located on the upper Coastal Plain physiographic region in Aiken, Barnwell, and Allendale Counties, South Carolina (33°0'-25'N, 81°25'-50'W). All individuals were collected under South Carolina Department of Natural Resources scientific research permit #G-02-09 and University of Georgia IACUC number A2002-10019-c2.

We randomly assigned individuals to one of two treatments: 1) subcutaneous implantation of tag and 2) a sham operation as a control in which individuals underwent the same procedure as treatment animals except no tag was implanted. Because we only captured 19 animals for the experiment, ten individuals were assigned to the treatment group and 9 to the control. We anesthetized animals in 30% ethanol (9) and recorded weight (g) for each individual. We sterilized all instruments and tags in 100% ethanol for at least half an hour prior to surgery. We inserted an 18-gauge catheter needle under the skin on the dorsal side of the animal, beginning at the anterior end of the animal, posterior to the neck. We inserted the catheter along the back, lateral to the spine, stopping before the pelvic bone. We removed the catheter from the animal and inserted a 0.1-g tag with two 11-mm antennae (total length 25.9 mm) into the hole created by the needle. We covered the tag with a thin layer of KY® liquid lubricant (Johnson & Johnson, New Brunswick, NJ) to reduce friction and facilitate a smooth entry into the cavity and then used tweezers to guide the tag until completely under the skin. We allowed animals to soak in water for an hour to aid in recovery from the effects of the ethanol solution and then placed individuals into 20-cm by 15-cm by 5-cm plastic containers with a damp paper towel. We fed each individual 20-30 termites every 7-14 days. We kept animals at 21°C and under natural light: dark period. We monitored individuals for 60 days, at the end of which they were weighed to determine if tags had any effects on individual body weight. We tested treatment effect on weight using a two-sample Wilcoxon rank sum test. We performed analysis with SAS software (10).

RESULTS

Detection distances for tags within canned sausages generally decreased with decreasing antenna length and depth of tag below the soil surface (Figure 2). Greatest detection distance was obtained from surface readings of tags having 9-mm antennae (144 cm). Tags with 7-mm antennae, however, could still be detected 30 cm below the soil. Tags located under small and large CWD were detected only when the transceiver was placed within 2.5 cm of the ground near the CWD.

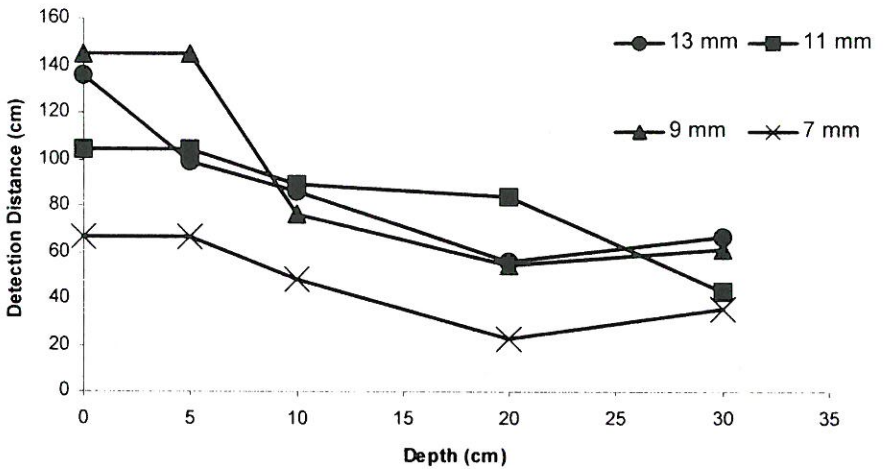


Figure 2. Detection range of reflector tags with varying antennae lengths ($n = 1$ each) inserted into canned sausages and buried at 4 depths below the soil surface.

Individuals generally recovered quickly following surgery with healing evident 2-5 days post-surgery. However, during the first 6 days following implantation, 3 individuals (2 treatment, 1 sham) died. Another treatment individual died 12 days after implantation. Retention rate of tags implanted in mole salamanders ranged from 14-60 days, with a mean (\pm SE) retention time of 42.5 days (\pm 6.5). Of the 7 surviving individuals implanted with tags, only 2 (29%) retained their tags for the duration of the study. The 5 individuals that lost their tags survived, healing rapidly following expulsion of the tag. Weight of individuals was not different between treatments ($Z = -0.18$, $P = 0.861$; Table I).

Table I. Mean body weight (\pm SE) before and after 60-day period (April 18, 2003 – June 16, 2003) in which mole salamanders (*Ambystoma talpoideum*) were given sham procedures or subcutaneously implanted reflector tags.

	Treatment (n)	Sham (n)	Z	P
Weight (g) at 0 Days	7.3 \pm 0.4 (7)	6.9 \pm 0.4 (8)	0.52	0.601
Weight (g) at 60 Days	7.0 \pm 0.5 (7)	6.7 \pm 0.4 (8)	0.29	0.771
Change in Weight (g)	0.3 \pm 0.1 (7)	0.2 \pm 0.1 (8)	-0.18	0.861

DISCUSSION

Mole salamanders burrow an average of 3-4 cm below the soil surface (1). Therefore the smallest tags in our study should be adequate for detecting individuals within the soil. Additionally, when tags were removed or fell out

of sausages, they were no longer detected. This characteristic of tags may prove beneficial in that expelled tags will not provide false signals that will mislead investigators.

We acknowledge that the level of mortality in our study is unacceptable and is likely attributable to the 30% ethanol used for anesthetic. Other researchers have experienced similar problems. Two marbled salamanders (*A. opacum*) died within the first two weeks following anesthetization with 2-phenoxy ethanol and passive integrated transponder (PIT) implantation (11). We encourage researchers to use alternative anesthetics in the future.

In our second experiment, the first signs of tag expulsion often occurred within 8 days of implantation. Bending of the body pushes the tag toward the posterior end, eventually applying pressure on the skin near the tail region. Other animals in which the tag was not pushed toward the tail region through normal body movement lost their tags when an ulcer formed in the area of the skin where the tag was applying pressure, and was eventually expelled through the ulcerated region. A similar response was observed for radioactive ^{182}Ta wire tags implanted into mole salamanders (12). Effects of tag expulsion likely caused discomfort to the animals in our study. In contrast, individuals that retained tags did not appear to experience any discomfort. They were observed to be as active as individuals without tags.

Our 60-day tag retention study indicates that use of subcutaneously implanted reflector tags on mole salamanders may not be an effective method for tag attachment without further improvement. Using only the largest individuals captured or smaller tags may have improved the 29% tag retention rate. Semlitsch (12) reported 83% of radioactive ^{182}Ta wire tags were expelled 100 days after implantation. Although we experienced poor tag retention, our tags were successfully used in another study where similar tags were retained for as long as 8 months (13). We recommend that future investigations focus on implantation procedures to improve retention rate of subcutaneously implanted reflector tags before this technique can be more widely applied in the field.

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