# Recommendations for Using the Subdermal Stitch Method to Attach External Transmitters on Snakes

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*Abstract.*—Snakes are a particularly neglected taxon within the field of wildlife management and conservation. Although research provides knowledge for evidence-based decision making, many factors hinder snake research. Challenging logistics regarding tracker attachment is one such factor. Radiotelemetry is a widely used method to track snakes, but internal implants require prohibitively expensive surgeries (two per snake), can result in potentially fatal infections, and are unsuitable for small snakes. Several methods for temporarily attaching external trackers to snake have largely failed. The subdermal stitch method for external transmitter attachment, however, appears to be a viable alternative, with fewer complications and injuries than the glue and glue-and-tape methods. We tested the efficacy of using the subdermal stitch method to attach radio-transmitters externally to Common Death Adders (*Acanthophis antarcticus*). In this pilot study, we tracked five individuals for 5–33 d (mean 20.8 d; median 21 d) on Magnetic Island, Queensland, Australia, in November and December 2018. We encountered multiple issues associated with the external attachment technique in death adders, including wounds to the tail (although none required veterinary services) and several entanglement hazards, one of which resulted in the death of one individual. We compare our experiences to previous studies, provide nine key recommendations that will guide future pilot studies to successfully test this technique, and call for more pilot studies to test this technique on snake species that range in ecology, morphology, and behavior.

*Key Words.*—*Acanthophis*; death adder; radio-tracking; radiotelemetry; snake tracking; subdermal stitch method; suture method; translocation

### INTRODUCTION

Snakes are a particularly neglected taxon within the field of wildlife management and conservation (Mullin and Seigel 2009). Multiple factors hinder our understanding of snake ecology and how to best manage human-snake interactions, including the cryptic nature of snakes, limited funding for snake research (Mullin and Seigel 2009), and the lack of cost-effective and practical methods to identify and track individuals. Several financial and logistical constraints limit snake research. Because they are limbless, snakes are unsuitable for conventional marking techniques, such as leg bands/ rings and transmitter collars. To overcome this barrier, many studies have used internal radio-transmitters to track snakes. These radio-tracking devices were originally force-fed into the snake during the 1970s and later surgically implanted into the body cavity for greater retention (Reinert et al. 1982; Whitaker et al. 2003; Croak et al. 2013; Marshall et al. 2019).

Despite acquiring important movement data previously impossible for snakes, implanting radiotransmitters is not without concern. Surgically implanting transmitter devices requires two invasive surgeries (one to insert and later to remove), which can be expensive and can cause infection (e.g., 60% infection rate in Eastern Massasaugas, Sistrurus catenatus catenatus; Lentini et al. 2011) that may lead to death (e.g., 11-28% mortality rate in Timber Rattlesnakes, Crotalus horridus, and Louisiana Pinesnakes, Pituophis melanoleucus ruthveni; Reinert and Cundall 1982; Rudolph et al. 1998). Also problematic is captivityrelated stress (DeGregorio et al. 2017), seasonal limitations for surgeries (Rudolph et al. 1998), and requiring up to 14 d to recover from surgery. In addition to these problems, because of the size of the currently available transmitter devices, surgical implantation is inappropriate for thin-bodied snakes, small snake species, or small individuals (e.g., juveniles; García-Aguayo 2008). Furthermore, some snake species can expel implanted transmitters intraperitoneally through the alimentary tract (Pearson et al. 2002).

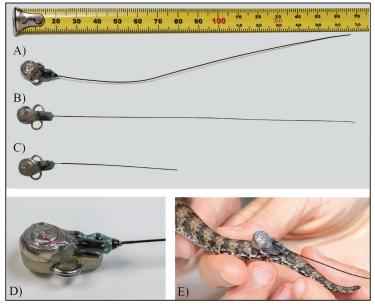
Another major concern for surgical implantations is financial cost. Previously, and currently in some countries (e.g., Thailand; Marshall et al. 2019), Australian researchers who were skilled personnel were permitted by animal ethics committees to perform surgeries themselves after veterinarian training (Grigg et al. 1989; Webb et al. 1997). Most Australian animal ethics committees now impose onto researchers a requirement to enlist the services of qualified veterinarians, regardless of a researcher having many years of experience in performing the surgeries on snakes (e.g., Croak et al. 2013). This requirement exists despite a low frequency of complications from surgeries performed by experienced researchers, and despite most veterinary surgeons lacking experience in surgical and anesthetic protocols for wildlife species (Anderson et al. 2006). We were quoted AU\$1,000/d by a qualified veterinarian for a maximum of four surgeries per day, with surgery, food, and accommodation expenses paid in addition to that amount. At this rate, implanting tracking devices into 10 snakes would cost a minimum of AU\$4,100 (excluding the cost of the devices). These prohibitively expensive costs greatly limit poorly funded snake research (Mullin and Seigel 2009), which could otherwise improve management and conservation of a species. Thus, although ethical treatment of animals is important, some animal ethics committees are being overly restrictive and not considering financial and practical constraints of research, thus hindering conservation efforts.

Further exacerbating the financial burden of veterinarian-performed surgeries is a recent (2018) change to the wildlife research permit legislation in Queensland, Australia. Queensland now restricts wildlife keeping for research purposes to less than 24 h. With this restriction in place, researchers can no longer spend weeks collecting and accumulating study animals in preparation for a short veterinarian visit. Rather, animal collection is restricted to within 24 h of a veterinarian being present to perform the surgeries, which in turn either greatly increases the cost of the project or constrains sample size and scientific rigor.

The risk of infection from at least two invasive surgeries per individual, coupled with the prohibitive cost of many days or weeks of veterinarian services, has led snake researchers to seek alternatives for tracking snakes, such as external attachments (Ciofi et al. 1991; Waddell et al. 2016; Riley et al. 2017; Robinson et al. 2018). Tape-and-glue or tape-only methods have been tested, resulting in varying degrees of skin abrasion, bleeding, and scale removal under the transmitter (García-Aguayo 2008; Wylie et al. 2011; Riley et al. 2017), and some trackers attached by these methods fall off or shed off prematurely (Wylie et al. 2011). A thread bobbin technique has been recently proposed for herpetofauna research (Waddell et al. 2016); however, it is greatly limited by: (1) duct tape losing effectiveness in wet conditions; (2) only 300 m of distance for tracking; (3) thread weakness (standard thread can snap); or (4) thread strength (stronger thread can restrict animal movement).

One method of attaching radio-transmitters that is gaining popularity is the subdermal stitch method. This method was first tested in 1991 on a colubrid snake native to Italy (Green Whip Snake, Coluber viridiflavus, now Hierophis viridiflavus, 200-450 g in body weight; Ciofi and Chelazzi 1991) and has since been used in three subsequent studies including the present pilot study (Riley et al. 2017; Wolfe et al. 2018). This method requires local anaesthetic and can be performed in approximately 15 min by a researcher who has been trained by a veterinarian. The procedure involves administering a local anaesthetic injection to two piercing points on the tail, then a 14-gauge needle (same size used to implant microchips into snakes, cats, or dogs) is used to make a small subdermal passage (piercing) under and perpendicular to the tail vertebrae. A small catheter tube is then passed through the needle, the needle is removed, and a suture thread is fed through the catheter tube to secure the tracker dorsally onto the tail (Fig. 1). Riley et al. (2017) reported no deaths and minimal injuries using the subdermal stich method on four captive Corn Snakes (Pantherophis guttatus) and four wild Eastern Massasaugas (> 100 g in body weight), and although most of the 10 Dugite (Pseudonaja affinis) snakes  $(530 \pm 202.7 \text{ [standard error] g})$  in Wolfe et al. (2018) perished, the authors state this was because of reasons besides the transmitter (five died from predation; two died from motor-vehicle strikes). Thus, the literature to date suggests the subdermal stich method for external radio-transmitter attachment to snakes appears to be a viable and practical method worth exploring further.

Death adders (Acanthophis spp.) are highly venomous (Broad et al. 1979) elapid snakes that resemble vipers (Viperidae) in body shape (stout body; triangular head) and foraging mode (ambush predator). There are currently seven species recognized in Australia and at least one recognized in Papua New Guinea (Wüster et al. 2005; Maddock et al. 2015), all of which possess a slender tail that is laterally compressed at the distal tip for caudal luring (Hagman et al. 2008) and terminates in a small tail spine. In this pilot study, we tested the use of the subdermal stitch method (Ciofi et al. 1991; Riley et al. 2017) to attach external radio-transmitters to five Common Death Adders (A. antarcticus; hereafter, death adders) on Magnetic Island, Queensland (QLD), Australia. In alignment with recommendations from Marshall and Strine (2021) regarding reporting all results regardless of outcome in herpetology, we report multiple animal welfare issues encountered with this technique for this species, including tail wounds and multiple entanglement hazards, one of which led to the death of a snake. We provide eight specific recommendations to improve the use of the subdermal stitch method for



**FIGURE 1**. Three slightly different, customized radio-transmitters: (A-C) 1.5 g, 9 mm wide × 7 mm deep × 20 mm long, with 14 cm long × 1 mm wide antennas from Advanced Telemetry Systems, Gold Coast, Queensland, Australia, model R1655). Views show the transmitter and button battery encased in a plastic resin, with galvanized attachment rings (D) for external attachment via the subdermal stitch method (Ciofi et al. 1991; Riley et al. 2017). These were attached to the dorsal region of a death adder tail (E). Transmitter B differs from A by a silicone dollop at the end of the whip antenna (designed to prevent fraying over time). Transmitter C is the shortened version of B to prevent the antenna from knotting on vegetation. (A, D, and E photographed by Elliot Budd; B and C by Christina Zdenek).

future snake research, and we call for more pilot studies using this technique on a wide range of snake species.

#### MATERIALS AND METHODS

We conducted a pilot study to determine if the subdermal stitch method to attach radio-transmitters was suitable to track wild death adders. To do this, we attached VHF radio-transmitters (Model R1655; 1.5 g, 9 mm wide  $\times$  7 mm deep  $\times$  20 mm long, with 14 cm long  $\times$  1 mm wide antennas from Advanced Telemetry Systems, Gold Coast, Queensland, Australia) on free-ranging death adders from 3 November 2018 to 9 December 2018 on Magnetic Island, Queensland, Australia. Transmitters did not exceed 3.5% the total body weight.

We attached transmitters (Fig. 1) following the supplementary files in Riley et al. (2017), including a slightly different transmitter placement on females versus males to avoid damaging the hemipenes. We pierced males at the  $15^{th}$  subcaudal scale posterior to the cloaca, whereas females were pierced at the  $5^{th}$  subcaudal scale. To avoid stress associated with probing to determine the sex of individuals, we visually determined sex via clear presence of hemipene bulges in male tails of this species, as well as by their longer tails (> 20% of body length; Maddock et al. 2015). Note that hemipenes are much longer in sea snakes than terrestrial snakes (Gillett et al. 2017). To externally attach the transmitters, we first used an insulin syringe to apply a

local anesthetic (bupivacaine, 1 mg/kg) to the proposed entry/exit points of the 14-gauge needle, then waited for a lack of motor response in the tail region. We then inserted a 14-gauge needle (Model #8881200573; Covidien Monoject, Minneapolis, Minnesota, USA) to make a small subdermal passage (piercing) under and perpendicular to the tail vertebrae just under the skin. Then, we passed a small catheter tube (Model #461206; Covidien Argyle, Minneapolis, Minnesota, USA) through the needle, removed the needle (leaving the catheter tube in place), and cut the tube to length. Finally, we fed a sterilized 2-0 silk suture thread through the catheter tube, fed the tube through the two lateral attachment rings on the transmitter, and tied the thread with a square knot to secure the transmitter dorsally onto the tail (Fig. 1). After the procedure was complete, we applied an antiseptic cream to the scales surrounding the two entry points of the needle to minimize the risk of infection. We only attached transmitters if the tail was large enough to safely pierce the tail (visually assessed by CZ who performed the procedures, as per training by reptile veterinarian Josh Llinas), and the transmitter did not exceed 3.5% the body weight. We released animals 48 h after the attachment procedure was complete to allow the anesthetic to wear off (4-12 h; Schumacher and Yelen 2006) and the piercing wounds to dry (i.e., form a crust of dried lymph).

We relocated death adders (hereafter, fixes) daily using a standard radio receiver (model TR-4; Telonics, Mesa, Arizona, USA) and a handheld VHF antenna

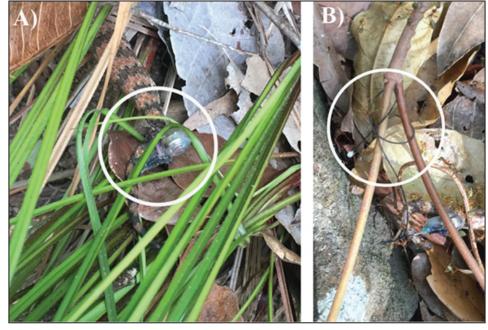
**TABLE 1.** Snake information for and issues sustained by five radio-tracked Common Death Adders (*Acanthophis antarcticus*) from Australia. Abbreviations are SVL = snout-vent length, F = female, and M = male. The asterisk (\*) indicates Green Ants (*Oecophylla smaragdina*).

ID	Weight (g)	SVL (mm)	Tail length (mm)	Category of snake	# days tracked	Issue sustained
AA01F	60.0	328	62	Resident	33	Divot wound
AA02M	82.0	410	81	Translocation	31	Scale abrasions
AA03F	47.6	285	75	Resident	5	Death by ants*
AA04F	76.2	385	75	Translocation	21	Tail constriction
AA05M	51.1	345	97	Resident	14	Scale abrasions

(model RA-23K, 148-154 MHz; Telonics). We acquired daily fixes on each snake from 3 November 2018 to 9 December 2018. Individual identification of death adders was possible via each transmitter emitting a different frequency signal in the 151 MHz range. During each fix, we uncovered the leaves if the snake was submerged under leaf-litter, visually assessed the health of the snake (particularly at the attachment point), noted if the snake was shedding, and recovered it with leaves. We did not perform statistical analyses because of insufficient sample size. Instead, we present visual and descriptive comparisons. Similar to Robinson et al. (2018), we subjectively categorized the severity of injuries as minor (constriction), mild (redness, flaking scales), moderate (cut through dermis, scabbing), or severe (deep laceration).

## RESULTS

We captured 10 wild Common Death Adders, five of which were large enough to include in the present pilot study. We attached external radio-transmitters to five snakes that had a mean body weight of  $63.4 \pm 18.40$  (standard deviation) g and mean tail length of  $78.0 \pm 12.70$  mm (Table 1). We encountered multiple animal welfare issues relating to the external transmitters (Table 1) during tracking of the animals (mean = 20.8 d; median = 21 d; range, 5–31 d). Four of the five death adders had varying degrees of damage or wounds that were only visible once the transmitters were removed at the end of the study. The fifth snake died five days after being released with the attached transmitter due to ant predation because of the transmitter whip-antenna becoming entangled in a native vine and entrapping the snake (Fig. 2). Additional

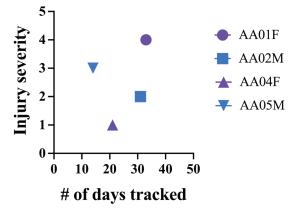


**FIGURE 2.** Two examples of entanglement issues. (A) A travelling Common Death Adder (*Acanthophis antarcticus*) was temporarily caught (white circle) on a high-tensile leaf of Blady Grass (*Imperata cylindrica*), which possesses finely-serrated margins and was the most common ground-cover in our study site. (B) A transmitter whip-antennae became knotted on a small native vine, resulting in the Common Death Adder carrying the transmitter being attacked and killed by Green Ants (*Oecophylla smaragdina*). (Photographed by Christina Zdenek).



FIGURE 3. Shed skin caught on the external radio-transmitter of (A) a male and (B, C) a female death adder. (C) Shed skin required manual removal to prevent catching on vegetation and inadvertent trapping of the snake. (Photographed by Christina Zdenek).

trapping hazards observed during the study included shed skin accumulating posteriorly on the body at the point of the transmitter (Fig. 3) and the transmitter alone getting caught on vegetation (Fig. 2). The number of tracking days did not appear to correlate with the injury level sustained by the snakes under the transmitter (Fig. 4). That is, while the snake that was tracked the longest had the most severe injuries from the transmitter, the snake that had the second-most severe injuries was tracked for the shortest amount of time, excluding the snake that



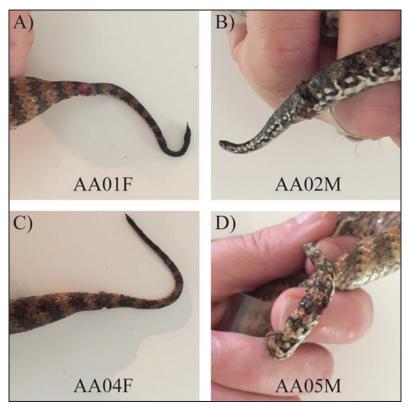
**FIGURE 4.** The severity of injury (1 = lowest; 4 = highest; details in Methods) sustained by radio-tracked Common Death Adders (*Acanthophis antarcticus*) compared to the length of time the snakes were tracked. Sex and duration of tracking were not visually correlated with injury severity. No injuries led to infection during the study, and none required veterinary care. AA03F was excluded because of death on day 5 of tracking. The abbreviations F = female and M = male in snake identification numbers.

died. We found various wounds on snakes caused by the transmitter (Fig. 5).

#### DISCUSSION

In this pilot study, we attached external radiotransmitters to five Common Death Adders using the subdermal stich method. We encountered multiple animal welfare issues relating to entrapment caused by the external transmitters. Each of these issues are described and discussed in detail below and compared to other studies (Appendix Table).

Shedding issue.-Shedding (i.e., sloughing) is a natural process that can occur because of body growth or in response to damage to scales and external parasites (Chris Hay, pers. obs.). In the death adders in our study, shed skin accumulated posteriorly at the point of the transmitter, creating a large and dangerous trapping hazard on their tail. An accumulation of shed skin was not reported in Riley et al. (2017), despite the wild snakes being tracked for over three months (20 June to 7 September 2013). This was possibly because of the rocky terrain and semi-aquatic marshes, bogs, and fens as opposed to the predominantly dry, leaf-litter terrain used by our snakes. Rocks probably provide a rubbing surface that the snakes could use to aid in breaking portions of the shed skin away in pieces, as opposed to a complete skin shedding off like a sock in soft leaf-litter. A semi-aquatic lifestyle may also help with shedding because soaking of skin softens it leading up to the shed.



**FIGURE 5.** Wounds sustained by death adders after (A) 21, (B) 33, (C) 31, and (D) 14 d of tracking on Magnetic Island, Queensland, Australia. Wounds include (A) divot wound from transmitter; (B, D) scale abrasion; and (C) constriction by catheter tube. Snakes were treated and held until wounds dried (72 h), then re-released (without transmitters attached) into a shady shelter site, away from Green Ant (*Oecophylla smaragdina*) nests, and nearest to the point of recent capture. (A and C photographed by Christina Zdenek and B and D by Chris Hay).

Although no snakes in our study died from shed skin accumulating at the transmitter, this is likely because of frequent (daily) fixes. That is, the two snakes that had transmitters attached for the longest period shed (with assistance), possibly because the transmitters were on long enough to induce a shed cycle. The third longest-tracked snake was approaching a shed when the transmitter was removed at the end of the study, and the two snakes tracked for the shortest periods (5 and 14 d) did not shed or begin the process. Therefore, longer studies might have a greater chance of the study snakes shedding and possibly encountering the issue of shed skin accumulating at the transmitter, thereby increasing entrapment risk.

Wolfe et al. (2018) used the subdermal stitch method to attach external radio transmitters to 10 Dugites (*Pseudonaja affinis*) in Perth, Western Australia, Australia. During the 2–49 d of tracking, the authors acquired fixes a minimum of once per week, and they did not observe any shedding events or signs of such for any of their snakes, so it is unknown whether clumping of shed skin would occur in this species or location. It is plausible, however, that shedding issues could have gone unnoticed and contributed to the death of four individuals by predation (possibly aided by entrapment events). The reason for this is because the shedding process is only visually obvious for 3–6 d prior to shedding, but not on every day leading up to the shed. At the beginning of the shed process, the skin color becomes duller (to the trained eye) and the eyes become opaque, but the eyes then clear to normal for about two days prior to the shed. As such, weekly fixes could miss the signs of a shedding process, whereas two fixes per week would increase the chances of observing a shed cycle and providing shedding assistance if required.

Aside from natural shedding cycles, the attachment of the transmitter itself may trigger a shedding cycle. This is because of unavoidable scale damage during the attachment process as well as abrasion under the transmitter and at the catheter tube insertion points caused by subsequent movement through the landscape. Indeed, Riley et al. (2017) observed an increased shed cycle frequency after significant damage to the skin when using both the tape-and-glue technique and the glue-only technique, and the authors had to aid in the shedding process with the tape-and-glue technique. Without an external factor such as scale damage inducing a shed cycle, we found natural shed cycles

**TABLE 2.** Shedding rate (mean  $\pm$  standard deviation) for four captive adult death adders: two Common Death Adders (*Acanthophis antarcticus*; AA) and two Rough-scaled Death Adders (*A. rugosus*; AR).

ID	Weeks between sheds	Number of sheds
AA07	$10.7 \pm 2.0$	12
AA09	$10.8 \pm 1.5$	12
AR15	$10.4 \pm 0.8$	12
AR18	$10.5 \pm 1.2$	12
Average	$10.6 \pm 1.4$	48 total sheds
Median	10.25	n/a

of adult captive death adders (*A. antarcticus* and *A. rugosus*, the Rough-scaled Death Adder, > 3 y of age) to be once every 10.6 ± 1.4 weeks (n = 48 sheds from four individuals; Table 2). Although the shed cycle of wild death adders is unknown, it is likely to be less frequent than captive individuals because of less regular large feedings.

Shed cycles may naturally occur more frequently in active foraging species compared to ambush species like Death Adders because of chemical crypsis in ambush snakes (Miller et al. 2015). Indeed, an adult (two-year-old) captive Coastal Taipan (*Oxyuranus scutellatus*) shed cycle was  $7.5 \pm 1.2$  weeks (n = 12 sheds from one individual; Barnett 1986), which is approximately three weeks quicker than adult (three years old) captive Common Death Adders. Moreover, unless shed cycles are always triggered by the transmitter attachment process, snakes with quicker shed cycles may be more at risk of entrapment because of the shedding process.

Entrapment issues.-From our experience, the greatest risk to using external trackers on snakes is entrapment, which can lead to exposure to environmental extremes, predators, or both. Not including the observed shedding entrapment discussed above, we observed transmitter entrapment by vegetation twice during the study, one of which resulted in the death of an animal due to predation by the abundant Weaver or Green Ant (Oecophylla smaragdina) at the study site. This particular incident was likely caused by the silicone dollop at the end of the whip antenna (designed to prevent fraying over time) getting caught in a vine junction, preventing forward movement of the snake. Given the knot and twists in the antenna present upon the discovery, it appears the snake managed to knot the short (145 mm) antenna. No further knot-induced entrapment events occurred after our mitigation measure of subsequently trimming all whip antennas in half.

From a practical perspective, it is important to note that shortening the antenna by 50% reduced the audible

distance of the radio-signal of the transmitter by about 50%, which may limit tracking species with large (> 500 m<sup>2</sup>) home-ranges. We report no obvious fraying of an antenna without the dollop, which was deployed for 33 d (this antenna was not fitted with a dollop by the manufacturer). The risk of entrapment will likely increase with larger leading edges of the transmitter (i.e., anterior transmitter depth when attached on the dorsal region of the tail). The attachment rings on our transmitters were adhered by the manufacturer below the battery, and this nearly doubled the original depth of the leading edge of our transmitters. Had the rings been cut in half and attached adjacent to the battery instead (or thin holes simply drilled through the resin), the transmitters would have been much more streamlined and probably less likely to catch on vegetation during forward movement. Although we did not measure the tail girth of our snakes at the point of transmitter attachment, it is clear from photos that the tail girth was around the same size or slightly larger than the transmitter depth at the leading edge. Given the inability of our snakes to overcome entrapment issues without assistance, we thus recommend ensuring at minimum a 2:1 ratio of tail girth (diameter at attachment site) to transmitter depth (at leading edge) and also width (at widest point at the battery). Additional resin in a streamlined fashion at the leading edge of the transmitter, as was used by Ciofi and Chelazzi (1991), may further reduce the risk of entrapment, although additional weight to the transmitter would have to be accounted for when choosing suitable snakes.

The risk of entrapment will change according to the habitat where the snakes are tracked. Generally, the more structurally and vegetatively dense a habitat, the greater the risk of entrapment in vegetation. For example, Madrid-Sotelo and García-Aguayo (2008) observed a temporary stop in forward movement of Mexican Vine Snakes (Oxybelis aeneus) with tape-and-glued external transmitters when the snakes passed through confluent branches, but not otherwise. Furthermore, grasses and small vines may be particularly problematic due to their small size and high tensile strength. Even within a habitat, microhabitat differences and chance can affect the number of entrapment events, which would explain the apparent lack of correlation between length of tracking and injury level sustained under the transmitter (due to the downward force upon the distal end of the transmitter produced by the attempted movement forward of the snake during entrapment). By contrast, we did not observe leaf litter, soil, or rocks to cause any entrapment issues in the present study.

The risk of entrapment will also change according to the snake species being tracked, particularly in relation to body weight and tail morphology. For example, although the girth of the transmitter plus the tail was less than the maximum diameter of the bodies our snakes, if this was not the case it would likely cause an additional trapping hazard. The foraging mode of the snake may also affect its risk of entrapment. Because they travel farther distances, active foragers may be more at risk of entrapment, but they may also have greater mobility and dexterity to overcome entrapment compared to sluggish, obligate ambush predators. Riley et al. (2017) reported no entrapment issues with Eastern Massasaugas (Sistrurus catenatus). The four snakes included in that study were all heavier (> 100 g) than the snakes included in our study (average 63.4 g), and the approximately 60% larger weight of the rattlesnakes may help explain the differences in how the two species fared with this technique. The percentage weight of the tracker compared with body mass may also contribute to outcomes: in Riley et al. (2017), the transmitter was never > 1.2% of the body mass of the snake, whereas in the current study our cut-off was 3.5% (which dropped the sample size of snakes in half: from 10 to five). Similarly, Wolfe et al. (2018) attached 14 g telemetry packages to snakes that weighed  $530 \pm 202.7$  g (2.6% of body mass on average); however, a high mortality rate was reported, as discussed above.

Tail morphology, particularly more muscle mass in the tail, is likely to allow a snake to better support the tracker (possibly leading to fewer wounds) and free itself from an entrapment situation. Rattlesnakes have robust tails capable of rapid rattling behavior, and the snakes also hold the rattle upwards when travelling forward, whereas all death adder species have short and skinny tails used for fine wriggling movements during caudal luring, and they do not lift their tails when travelling forward. Although we were unable to observe caudal luring behavior in any of our snakes, nor did we measure the weight of our snakes over the relatively short tracking period (5-33 d), we do not suspect the snakes had any difficulty maintaining natural caudal luring ability. This is because only the posterior 20 mm of the tail wriggles during caudal luring, and the transmitter was much farther (about 30 mm) anterior from the tail tip. Thus, body size and shape (e.g., abundance and distribution of muscle) is likely to play a major role in determining the success of the subdermal stitch technique for a given species, with larger bodies and relatively larger tails seemingly favored.

Arboreal and fossorial behavior may also alter entrapment frequencies. The Brown Tree Snake (*Boiga irregularis*), a climbing species, was found to use the tail less while the transmitter was attached via the tapeglue-tape method, but the transmitter did not appear to affect overall mobility or feeding behavior (Robinson et al. 2018). Fossorial species may be more sensitive to the girth (depth) of the transmitter because the snakes use small holes in the ground, so the discussed options for reducing transmitter depth and making it more streamlined would presumably be particularly prudent for fossorial species.

Home-range size, which is contingent on foraging mode (i.e., ambush vs. active) is another potential major factor in the risk of entrapment. Similarly, mode of movement (e.g., rectilinear, sidewinding, concertina) should be considered. Because of the limited number of snake species used with the subdermal stitch method to date (five including our study), however, it is unknown whether home-range sizes and mode of movement affect the frequency and duration of entrapment events. Regardless of the species, it is possible that a small amount of glue to fix only the leading edge of the transmitter to the body may mitigate the risk of grass and other vegetation becoming caught between the transmitter and the body at the leading edge of the transmitter. Gluing the whole transmitter as the means for attachment has been recommended against because this was shown to cause skin irritation (Robinson et al. 2018) and bleeding (Riley et al. 2017).

Wound issues.- The initial piercing wound caused by the attachment procedure creates two small (2 mm diameter) wounds: one each at the entry and exit point of the needle. We were unable to determine the length of time for closure of these wounds to occur because the wounds were visually blocked by the catheter tube. We found that a 48-h waiting period after the procedure was required to allow the wound to dry enough (i.e., form a crust of dried lymph) so as to not attract ant predators (Green Ants), as one snake that we attempted to release at the 24-h mark attracted Green Ants within 10 min of release. Thus, although 24 h for wound closure was deemed sufficient in Riley et al. (2017) and Wolfe et al. (2018), we deemed the 48-h waiting period extremely important in order to not attract Green Ants to the piercing wound sites.

Wounds sustained by snakes due to external transmitters over time can be caused by normal rubbing, but they can also be exacerbated by entrapment events. We witnessed several such events to result in additional force being exerted upon the body by the distal end of the transmitter. Indeed, during daily early morning fixes, the only snake to never be observed entrapped (AA04F) was the snake with minor injuries (constriction only, due to the catheter tube). Despite close observation of the tail region during daily fixes, wounds under the transmitter were only visible once we removed the transmitters at the end of the study. These wounds required a holding period of 72 h to dry. No injuries led to infection during the study, and none required veterinary care. Long-term recovery of the small wounds could not be assessed because the snakes could not be relocated in the environment without their transmitter.

The wounds observed in the current study varied in severity between snakes but did not appear to be correlated with length of time in the field. Types of wounds included constriction by the catheter tube, scale abrasions, and a divot wound, which was the most serious wound observed but still did not require veterinary attention. This wound was likely caused by downward force upon the distal end of the transmitter during entrapment event struggles. Excluding the divot wound, all other wounds were less severe than the wounds sustained during the pilot study by Riley et al. (2017) using the tape-and-glue method for attachment, that is, scale malformations, minor and severe bleeding, and removal of about 10 adjacent scales, although no injuries were observed in the captive pilot study within that study. Interestingly, wounds were not reported by Wolfe et al. (2018). Furthermore, all wounds sustained by our snakes were still superficial and minor, albeit more unpredictable, compared to two invasive surgeries required for surgical implantation of devices. Thus, from an animal welfare perspective, the wounds sustained by our snakes using the subdermal stitch method were less severe than those observed with both the tape-and-glue external attachment method and especially compared to the surgical implantation method.

Nine key recommendations.—While more data and greater sample sizes are preferred before providing conclusions and making recommendations, depauperate reptile funding often precludes this luxury. With what knowledge we do have, we recommend the following measures be considered in future studies that use the subdermal stitch method for attaching transmitters on snakes: (1) Conduct a pilot study prior to using the subdermal stitch method, especially for small elapid snakes because they may not have the strength to overcome entrapment situations. This test period in the field for each species and habitat would allow researchers to gauge the entrapment risks involved, determine the holding time required for the piercing wounds to dry in order to not attract predators, and determine the rate of fixes required to ensure safety but limit altering snake behavior (but see #8 below). (2) Consider vegetation type and abundance prior to using this method, as certain plant types (e.g., highly tensile grasses) and structures (e.g., dense vegetation) can increase the risk of entrapment. (3) Give careful attention to the customized attachment options (i.e., rings) on the transmitter, as this can double the depth of the leading edge of the transmitter and increase the chance of entrapment events. The shallower the depth of the transmitter and the more streamlined it is, the less likely it will catch on vegetation. (4) To reduce the depth of the transmitter and thus the size of the leading

edge, either drill holes into the resin for attachment points, or cut the attachment rings in half and place them adjacent to the battery, not under the battery, ensuring the inner radius of the half-ring can still accommodate the catheter tube for attachment purposes. (5) Consider adding a customized rounded anterior portion to the transmitter (Ciofi and Chelazzi, 1991) to reduce the risk of entrapment. (6) Consider using a minimum tail-totracker ratio for a threshold cut-off of snakes suitable for this method. We recommend at minimum a 2:1 ratio of tail girth (diameter at attachment site) to transmitter depth (at leading edge). (7) Consider a minimum body weight limit so the transmitter weighs no more than 2% the body mass of the snake. (8) Remove any silicone dollop from the whip antennas of the transmitters prior to field deployment. This dollop otherwise may cause entrapment. (9) Determine the rate of fixes required to ensure safety but limit altering snake behavior. We recommend daily fixes for vulnerable periods (e.g., initial release and shedding periods) and once every 1-3 d otherwise. This will help ensure safety of the snake and enable shedding events to be noted and given aid if required. Unless under cover, fixes can be completed using binoculars to avoid stressing the animal and altering behavior.

*Conclusion*.—Three prior studies across four species reported largely positive results from the use of the subdermal stitch method for tracking snakes (Ciofi et al. 1991; Riley et al. 2017; Wolfe et al. 2018). Our pilot study on Common Death Adders, however, encountered multiple issues with the external transmitter relating to habitat, small snake size, and limited muscle strength. While these issues led to minor tail wounds that were less severe than other external transmitter attachment methods reported in the literature, several entanglement hazards raised concern, one of which resulted in the death of an individual. As such, with the current size of transmitters, we caution against the use of this technique for small snakes in particular; however, due to the limited tracking method options available for small snakes, issues relating to other tracking methods, and the many aforementioned limitations to snake research, we also call for more pilot studies to test the subdermal stitch for other snake species. Ideally, future pilot studies should include species with a diversity of ecological niches, morphologies, modes of movement, and behaviors.

Acknowledgments.—Funding was provided by National Geographic (90%) and Australian Geographic (10%). We thank Dr. Josh Llinas (Unusual Pet Vets Jindalee) for training CZ on microchipping and the subdermal stitch method procedure and equipment consultation; Dr. Julia Riley for initial consultation; Amaroo On Mandalay for accommodation for our team; SeaLink for in-kind support via ferry and barge tickets; Elliot Budd for a car loan for roadcruising; Lyn Beard for radio-tracking training for CZ and CH and loaning tracking equipment; and the following volunteers for search help: Elliot Budd, Justin Wright, and Daniel Bromley. This work was approved by the Queensland Department of Environment and Science for non-protected areas (Permit No. WA0008981) and the Department of National Parks, Racing, and Sport (Permit No. PTU18-001289) for protected areas. Animal ethics approval was received from the Animal Ethics Committee of The University of Queensland (Approval No. SBS/ANU/234/18).

## LITERATURE CITED

- Anderson, C.D., and M. Talcott. 2006. Clinical practice versus field surgery: a discussion of the regulations and logistics of implanting radiotransmitters in snakes. Wildlife Society Bulletin 34:1470–1471.
- Barnett, B. 1986. The Taipan (*Oxyuranus scutellatus*) in captivity. Thylacinus 11:9–19.
- Broad, A.J., S.K. Sutherland, and A.R. Coulter. 1979. The lethality in mice of dangerous Australian and other snake venom. Toxicon 17:661–664.
- Ciofi, C., and G. Chelazzi. 1991. Radiotracking of *Coluber viridiflavus* using external transmitters. Journal of Herpetology 25:37–40.
- Croak, B.M., M.S. Crowther, J.K. Webb, and R. Shine. 2013. Movements and habitat use of an endangered snake, *Hoplocephalus bungaroides* (Elapidae): Implications for conservation. PLoS ONE 8:1–10. https://doi.org/10.1371/journal.pone.0061711.
- Degregorio, B.A., J.H. Sperry, T.D. Tuberville, and P.J. Weatherhead. 2017. Translocating ratsnakes: does enrichment offset negative effects of time in captivity? Wildlife Research 44:438–448.
- García-Aguayo, A. 2008. A simple method for externally attaching radio-transmitters to snakes. North-western Journal of Zoology 4:335–338.
- Gillett, A.K., R. Ploeg, M. Flint, and P.C. Mills. 2017. Postmortem examination of Australian sea snakes (Hydrophiinae): anatomy and common pathologic conditions. Journal of Veterinary Diagnostic Investigation 29:593–611.
- Grigg, G.C., L.A. Beard, and M.L. Augee.1989. Hibernation in a monotreme, the Echidna (*Tachyglossus aculeatus*). Comparative Biochemistry and Physiology - Part A: Physiology 92:609–612.
- Hagman, M., B.L. Phillips, and R. Shine. 2008. Tails of enticement: caudal luring by an ambushforaging snake (*Acanthophis praelongus*, Elapidae). Functional Ecology 22:1134–1139.
- Lentini, A.M., G.J. Crawshaw, L.E. Licht, and D.J. McLelland,. 2011. Pathologic and hematologic

responses to surgically implanted transmitters in Eastern Massasauga Rattlesnakes (*Sistrurus catenatus catenatus*). Journal of Wildlife Diseases 47:107–125.

- Maddock, S.T., R.J. Ellis, P. Doughty, L.A. Smith, and W. Wuster. 2015. A new species of death adder (*Acanthophis*: Serpentes: Elapidae) from northwestern Australia. Zootaxa 4007:301–326.
- Marshall, B.M. and C.T. Strine. 2021. Make like a glass frog: in support of increased transparency in herpetology. Herpetological Journal 31:35–45.
- Marshall, B.M., C.T. Strine, M.D. Jones, T. Silva, I. Artchawakom, P. Suwanwaree, and M. Goode. 2019. Space fit for a king: spatial ecology of King Cobras (*Ophiophagus hannah*) in Sakaerat Biosphere Reserve, Northeastern Thailand. Amphibia-Reptilia 40:163–178.
- Miller AK, B. Maritz, S. McKay, X. Glaudas, G.J. Alexander. 2015. An ambusher's arsenal: chemical crypsis in the PuffAdder (*Bitis arietans*). Proceedings of the Royal Society B - Biological Sciences 282:1– 8.
- Mullin, S.J., and R.A Seigel. 2009. Snakes: Ecology and Conservation. Cornell University Press, Ithaca, New York, USA.
- Pearson, D.J., and R. Shine. 2002. Expulsion of intraperitoneally-implanted radiotransmitters by Australian pythons. Herpetological Review 33:261– 263.
- Reinert, H.K., and D. Cundall. 1982. An improved surgical implantation method for radio-tracking snakes. Copeia 1982:702–705.
- Riley, J.L., J.H. Baxter-Gilbert, and J.D. Litzgus. 2017. A comparison of three external transmitter attachment methods for snakes. Wildlife Society Bulletin 41:132–139.
- Robinson, C., M.C. Viernes, R.N. Reed, A. Yackel Adams, and M.G. Nafus. 2018. Assessment of two external transmitter attachment methods for *Bioga irregularis* (Brown Treesnake). Herpetological Review 49:32–34.
- Rudolph, D.C., S.J. Burgdorf, R.R. Schaefer, R.N. Conner, and R.T. Zappalorti. 1998. Snake mortality associated with late season radio-transmitter implantation. Herpetological Review 29:155–156.
- Schumacher, J., and T. Yelen. 2006. Anesthesia and analgesia. Pp. 442–452 *In* Reptile Medicine and Surgery. 2<sup>nd</sup> Edition. Mader, D.R. (Ed.). Saunders Elsevier, St. Louis, Missouri, USA.
- Waddell, E., A.Whitworth, and R. MacLeod. 2016. A first test of the thread bobbin tracking technique as a method for studying the ecology of herpetofauna in a tropical rainforest. Herpetological Conservation and Biology 11:61–71.
- Webb, J.K., and R. Shine. 1997. Out on a limb:

conservation implications of tree-hollow use by a threatened snake species (*Hoplocephalus bungaroides*: Serpentes, Elapidae). Biological Conservation 81:21–33.

- Whitaker, P.B., and R. Shine. 2003. A radiotelemetric study of movements and shelter-site selection by free-ranging Brownsnakes (*Pseudonaja textilis*, Elapidae). Herpetological Monographs 17:130–144.
- Wolfe, A.K., P.A. Fleming, and P.W. Bateman. 2018. Impacts of translocation on a large urban-adapted venomous snake. Wildlife Research 45:316–324.
- Wüster W., A. J. Dumbrell, C. Hay, C. E. Pook, D.J. Williams, B.G. Fry. 2005. Snakes across the strait: trans-Torresian phylogeographic relationships in three genera of Australasian snakes (Serpentes: Elapidae: Acanthophis, Oxyuranus, and Pseudechis). Molecular Phylogenetics and Evolution 34:1–14.
- Wylie, G.D., J.J. Smith, M. Amarello, and M.L. Casazza. 2011. A taping method for external transmitter attachment on aquatic snakes. Herpetological Review 42:187–191.



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Sheries	Average body	Tracker weight (% hodv mass)	No. animals tracked	Duration of tracking	Min. rate of fixes	Foraging	Micro-habitat	Iniiries	No. deaths due to transmitter	Entrapment	Shedding issues?	Study
Green Whip Snake	313 g (m); 241 g (f)	0.9-1.0 %	=	5 mo	5/d	active forager	unknown ('10km north of Florence, Italy')	ou	0	no	ou	Ciofi & Chelazzi, 1991
Dugite	$530 \pm 202.7$ g	2.6 %	10	249 d	1/week	active forager	urban	ou	0	no	ou	Wolfe et al., 2018
Eastern Massasauga	> 100 g	1.2 %	4	2.5 mo	2–3/ week	ambush predator	rocky terrain and semi- aquatic marsh/ bogs/fens	constriction; some abrasion of scales under transmitter	0	no (brief snags on debris)	ou	Riley et al., 2017
Corn Snake	> 100 g	1.2 %	4	2 mo	2–3/ week	active forager	indoor obstacle course	ou	0	оп	no	Riley et al., 2017
Common Death Adder	$63.4 \pm 15.2 \text{ g}$	3.5 %	ŝ	5–33 d	1/d	ambush predator	leaf-litter	constriction; scale abrasion; divor wound	-	yes	yes	this study

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