

## Radio Transmitter Implantation and Movement in the Wild Timber Rattlesnake (*Crotalus horridus*)

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**ABSTRACT:** Radiotelemetry transmitters have become critical to studies of wildlife ecology. However, little is known about how transmitter implantation surgery affects the mobility of some species, including the timber rattlesnake, *Crotalus horridus*. Tracking snake movement can provide insights into the effects of transmitter implantation. During 2007–11, 71 radio transmitters were surgically implanted intracoelomically in 47 timber rattlesnakes. Over 20 of these snakes underwent surgery at least twice in 5 yr to replace old transmitters. Surgeries were performed under general anesthesia with a local nerve block at the site of implantation, 20 cm cranial to the cloaca. Snakes were also administered postsurgical meloxicam and enrofloxacin every 24 h for three doses. Two to five days after surgery, snakes were released at their original locations and radiotracked regularly during the active seasons (April–October 2007–11). Average daily movement data (distance traveled) were compiled for each snake. Snakes undergoing transmitter surgery in a given year did not differ significantly in distance traveled compared to snakes that had transmitters but did not have surgery in that year. Distance traveled for each snake did not differ before or after surgery or between weeks 1 and 2 postsurgery, indicating that the transmitter implantation did not alter snake movement.

**Key words:** *Crotalus horridus*, GIS, meloxicam, NSAID, radio transmitter.

Use of radiotelemetry devices in ecologic studies provides insight into wildlife biology and behavior, but transmitter application or implantation can be associated with fitness costs including increased energy expenditure (Godfrey et al. 2003), decreased mobility (Knapp and Abarca 2009), diminished reproductive success (Reed et al. 2005), and death

(Theuerkauf et al. 2007). In reptiles, intracoelomic implantation of radio transmitters is associated with additional surgical risks, including pain, inflammation, and infection (Rudolph et al. 1998; Ujvari and Korsos 2000; Lentini et al. 2011).

To characterize the perioperative movement of transmitter-implanted wild timber rattlesnakes (*Crotalus horridus*), we tracked their movements after transmitter implantation. Specifically, we addressed three questions:

1. Does movement vary in snakes that undergo surgery vs. snakes that do not undergo surgery in a given year?
2. Does average distance traveled per day vary pre- and postsurgery?
3. Does average distance traveled per day vary over time (week 1 vs. week 2) postsurgery?

We tracked timber rattlesnakes ( $n=47$ ; 24 females, 23 males; 11 females were gravid over the 5-yr study) in the Morgan-Monroe and Yellowwood State Forests between April and November 2007–11. Snakes were initially located as part of a larger study (Swihart et al. 2013; MacGowan et al. 2017) with the use of meandering transects and opportunistic encounters. Once found, snake sex, length, and weight (measured on a spring scale, Pesola®, Schindellegi, Switzerland) were recorded and the capture location was documented with the use of a handheld GPS device (Garmin Ltd., Olathe, Kansas, USA) (Table 1). Snakes were transported to the Indiana Department of

TABLE 1. Average weight and snout–vent length (SVL) of radio-tracked timber rattlesnakes (*Crotalus horridus*) in southern Indiana, USA during 2007–11.

Sex	Weight (g)		SVL (cm)	
	Mean (SD)	Range	Mean (SD)	Range
Female	752.7 (289.6)	260–2,365	97.4 (10.2)	72–116
Male	1,680.2 (550.5)	325–2,770	120 (13.3)	76–151
Total	1,228.5 (641.4)	260–2,770	109 (16.4)	72–151

Natural Resources office (Bloomington, Indiana, USA), and housed in secure 76-L glass aquariums for 1–5 d before surgery. We performed 71 transmitter implantation surgeries; 90% of them ( $n=64$ ) in May, June, or July.

For surgery, the anterior third of the snake was placed in a clear restraining tube connected to an isoflurane vaporizer (IsoFlo, Abbott Laboratories, Chicago, Illinois, USA). Intracoelomic radio transmitter (Holohil Systems Ltd., Carp, Ontario, Canada; models AI-2T or SI-2T) implantation was performed under general anesthesia with the use of aseptic technique and under surgical protocols described previously (Reinert and Cundall 1982; Frye 1991; Martin-Bashore and Bashore 2001; Anderson et al. 2005). Transmitter weight (9 g, 13.5 g, or 25 g) did not exceed 5% body weight, and all but one transmitter (with a 1-yr battery life) had battery lives of 2–3 yr. Transmitters were placed 20 cm cranial to the cloaca and caudal to the lung of the snakes. Transmitter antenna wires (20 cm) were tunneled subcutaneously and directed caudally with the use of a polypropylene urinary catheter as a stylet to limit tissue trauma. During closure of the body wall, the antenna wire was secured with one interrupted suture (Monocryl Plus, Ethicon, Sommerville, New Jersey, USA). We performed 71 transmitter implantation surgeries; 24 of these were transmitter replacement surgeries in previously implanted snakes (20 snakes had two surgeries, two had three).

Before surgery, a regional block was performed with mepivacaine (Carbocaine-V, 20mg/mL, Pfizer-Pharmacia & Upjohn Com-

pany, New York, New York, USA; 2–6 mg/kg subcutaneously divided between three intercostal spaces, 20 cm cranial to the cloaca) at the implantation site (Wellehan et al. 2006). Snakes received three postsurgical injections of meloxicam (Metacam, 5 mg/mL, Boehringer Ingelheim, Fort Dodge, Iowa, USA; 0.2 mg/kg) and enrofloxacin (Baytril, 22.7 mg/mL, Bayer Healthcare LLC, Shawnee Mission, Kansas, USA; 5–10 mg/kg) intramuscularly every 24 h. Both injections were adjacent to the dorsal spinous processes in the anterior half of the snake, but caudal to the restraining tube.

Snakes were released at their original capture locations after 2–5 d of postsurgical monitoring and tracked 2–3 times weekly until they returned to their hibernacula in the fall. Tracking resumed the following spring and each year thereafter through 2011. Morbidities and mortalities from surgery in this study were rare, with only one overwintering mortality (S.T. unpubl. data). This study was approved by the Purdue Animal Care and Use Committee (PACUC 07-038).

Euclidian distances between GPS locations for each snake were calculated with the use of the HRT extension in ArcGIS (Home Range Tools for ArcGIS, version 1.1; ESRI ArcGIS Desktop v. 9.3.1, Redlands, California, USA). Average Euclidian distances were divided by the average number of days between locations to calculate steplength. Steplengths were summed to calculate total weekly, monthly, and yearly distance traveled for each snake.

To compare distances traveled between groups, repeated-measures linear mixed models with post hoc LSMEANS pairwise comparisons were run in SAS 9.3 (SAS Institute

Inc., Cary, North Carolina, USA). Sex and surgery were fixed effects; month, year, and animal ID were random effects. Because there was no significant difference between distance traveled by gravid and nongravid females ( $t_{136}=1.27$ ,  $P=0.21$ ), we combined both categories as “females” in subsequent analyses. Steplength values were log transformed to meet the assumptions of equal variances and normal distribution.

There was no significant difference in steplength between snakes that did or did not undergo surgery in a given year ( $n=47$ ; surgery mean: 34.9 m,  $SE=1.8$ ; no surgery: 34.5 m,  $SE=1.6$ ;  $F_{1,501}=0.09$ ;  $P=0.76$ ). Steplength by individual snakes did not vary in the 14 d before or after surgery ( $n=22$ ;  $F_{1,41}=0.18$ ;  $P=0.67$ ). Sex did not have a significant effect in this model ( $F_{1,41}=3.68$ ;  $P=0.062$ ). Steplength for males ( $n=6$ ) decreased slightly but not significantly from a presurgery mean of 45.9 m ( $SE=23.6$ ) to 42.2 m ( $SE=7.8$ ) post-surgery. For females ( $n=16$ ), presurgery steplength (22.2 m,  $SE=5.1$ ) was almost identical to postsurgery (21.1 m,  $SE=4.8$ ). In the model comparing distance traveled in weeks 1 and 2 postsurgery, sex was significant ( $F_{1,45}=7.60$ ;  $P=0.008$ ; male  $n=9$ , female  $n=15$ ); however, average total steplength by week (week 1 vs. 2 postsurgery) was not ( $F_{1,45}=4.00$ ;  $P=0.052$ ; week 1 male: 43.1 m,  $SE=10.7$ , female: 20.8 m,  $SE=7.5$ ; week 2 male: 53.0 m,  $SE=9.2$ , female: 35.1 m,  $SE=10.2$ ). Because all snakes had transmitters implanted, we could not determine how transmitters affect snake movement compared to snakes without transmitters; we could only make observations on whether movement of transmitter-implanted snakes was altered perioperatively.

Though we cannot decisively conclude that our analgesic protocol achieved therapeutic concentrations and mitigated pain, our results suggest that transmitter implantation surgery in concert with analgesics does not alter snake movement in the short term (14 d postsurgery) or long term (over the season). In reptiles, altered or decreased vagility is commonly used as a behavioral assessment of pain (Mosley 2011; Sladky 2014). Although pain is not well understood in reptiles;

anatomically and physiologically, reptiles maintain central nervous system pathways, peripheral nociceptors, endogenous opioids, and opioid receptors consistent with processing noxious stimuli (Machin 2001; Mosley 2009, 2011; Sladky 2014). As such, reptiles may be good candidates for analgesics.

The dose of meloxicam we used (0.2 mg/kg) was consistent with other studies on reptile analgesia and with current recommendations in veterinary formularies for alleviation of pain (Mader 2006; Wellehan et al. 2006; Ramsey 2008; Divers et al. 2010; Sladky and Mans 2012; Carpenter 2013). Pharmacokinetics and pharmacodynamics of meloxicam are poorly understood in snakes and were not examined in this study, but have been reported for similar dosing in other reptiles including green iguanas, *Iguana iguana* (Divers et al. 2010); red-eared sliders, *Trachemys scripta elegans* (Uney et al. 2016); and loggerhead sea turtles, *Caretta caretta* (Lai et al. 2015). Interpreting the therapeutic value of various meloxicam plasma concentrations, however, still remains challenging. Olesen et al. (2008) found that a dose of 0.3 mg/kg meloxicam had no apparent analgesic effects on ball pythons, *Python regius*, and recent clinical practitioners report routine use of meloxicam at much higher doses in reptiles (0.5–1.0 mg/kg every 24 h) without negative effects (Meredith 2015). Although much remains to be learned about reptile analgesia, it is particularly important to consider in wild snake transmitter implantation as the potential for pain to alter postsurgical activity can have serious implications on prey capture and predator escape.

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#### LITERATURE CITED

- Anderson NL, Hetherington TE, Coupe B, Perry G, Williams JB, Lehman J. 2005. Thermoregulation in a nocturnal, tropical, arboreal snake. *J Herpetol* 39:82–90.
- Carpenter JW. 2013. *Exotic animal formulary*. 4th Ed. Saunders Elsevier, St. Louis, Missouri, 724 pp.
- Divers SJ, Papich M, McBride M, Stedman NL, Perpnan D, Koch TF, Hernandez SM, Barron GH, Pethel M, Budsberg SC. 2010. Pharmacokinetics of meloxicam following intravenous and oral administration in green iguanas (*Iguana iguana*). *Am J Vet Res* 71:1277–1283.
- Frye F. 1991. *Biomedical and surgical aspects of captive reptile husbandry*. Krieger Publishing Company, Malabar, Florida, 712 pp.
- Godfrey JD, Bryant DM, Williams MJ. 2003. Radiotelemetry increases free-living energy costs in the endangered Takahe *Porphyrio mantelli*. *Biol Conserv* 114:35–38.
- Knapp CR, Abarca JG. 2009. Effects of radio transmitter burdening on locomotorability and survival of iguana hatchlings. *Herpetologica* 65:363–372.
- Lai OR, Bello AVFD, Soloperto S, Freggi D, Marzano G, Cavaliere L, Crescenzo G. 2015. Pharmacokinetic behavior of meloxicam in loggerhead sea turtles (*Caretta caretta*) after intramuscular and intravenous administration. *J Wildl Dis* 51:509–512.
- Lentini AM, Crawshaw GJ, Licht LE, McLelland DJ. 2011. Pathologic and hematologic responses to surgically implanted transmitters in eastern massasauga rattlesnakes (*Sistrurus catenatus catenatus*). *J Wildl Dis* 47:107–125.
- MacGowan BJ, Currylow AF, MacNeil J. 2017. Short-term responses of timber rattlesnakes (*Crotalus horridus*) to even-aged timber harvests in Indiana. *For Ecol Manage* 387:30–36.
- Machin KL. 2001. Fish, amphibian, and reptile analgesia. *Vet Clin North Am Exot Anim Pract* 4:19–33.
- Mader DR, editor. 2006. *Reptile medicine and surgery*. 2nd Ed. Saunders Elsevier, St. Louis, Missouri, 1264 pp.
- Martin-Bashore TE, Bashore TL. 2001. *Field techniques for surgically implanting radio transmitters in venomous snakes*. [http://www.nmfwa.net/uploads/documents/Herp\\_Slideshow.pdf](http://www.nmfwa.net/uploads/documents/Herp_Slideshow.pdf). Accessed December 2016.
- Meredith A, editor. 2015. *Small animal formulary: Part B: Exotic Pets*. 9th Ed. British Small Animal Veterinary Association, Quengeley, Gloucester, England, 352 pp.
- Mosley C. 2009. Clinical approaches to analgesia in reptiles. In: *Handbook of veterinary pain management*, Gaynor J, Muir WW, editors. Mosby Elsevier, St. Louis, Missouri, pp. 481–493.
- Mosley C. 2011. Pain and nociception in reptiles. *Vet Clin North Am Exot Anim Pract* 14:45–60.
- Olesen MG, Bertelsen MF, Pery SF, Wang T. 2008. Effects of preoperative administration of butorphanol or meloxicam on physiologic responses to surgery in ball pythons. *J Am Vet Med Assoc* 233:1883–1888.
- Ramsey I, editor. 2008. *Small animal formulary*. 6th Ed. British Small Animal Veterinary Association, Quengeley, Gloucester, England, 488 pp.
- Reed ET, Gilles G, Roger P. 2005. Effects of neck bands on reproduction and survival of female Greater Snow Geese. *J Wildl Manage* 69:91–100.
- Reinert HK, Cundall D. 1982. An improved surgical implantation method for radio-tracking snakes. *Copeia* 1982:702–705.
- Rudolph DC, Burgdorf SJ, Schaefer RR, Conner RN, Zappalorth RT. 1998. Snake mortality associated with late season radio-transmitter implantation. *Herpetol Rev* 29:155–156.
- Sladky KK. 2014. Analgesia. In: *Current therapy in reptile medicine and surgery*, Mader DR, Divers SJ, editors. Elsevier Saunders, St. Louis, Missouri, pp. 217–228.
- Sladky KK, Mans C. 2012. Clinical analgesia in reptiles. *J Exot Pet Med* 21:158–167.
- Swihart RK, Saunders MR, Kalb RA, Haulton GS, Charles H. 2013. *The hardwood ecosystem experiment: A framework for studying responses to forest management*. General Technical Report NRS-P-108US. Department of Agriculture, Forest Service, Newton Square, Pennsylvania, 349 pp. <https://www.fs.fed.us/nrs/pubs/gtr/gtr-nrs-p-108.pdf>. Accessed December 2016.
- Theuerkauf J, Rouys S, Chatreau C. 2007. Mortality of radio-tracked wild rats in relation to transmitter weight and resilience of transmitters in relation to their design. *J R Soc N Z* 37:85–90.
- Ujvari B, Korsos Z. 2000. Use of radiotelemetry on snakes: A review. *Acta Zool Acad Sci Hung* 46:115–146.
- Uney K, Altan F, Aboubakr M, Cetin G, Dik B. 2016. Pharmacokinetics of meloxicam in red-eared slider

turtles (*Trachemys scripta elegans*) after single intravenous and intramuscular injections. *Am J Vet Res* 77:439–444.

Wellehan JF, Gunkel CI, Kledzik D, Robertson SA, Heard DJ. 2006. Use of a nerve locator to facilitate

administration of mandibular nerve blocks in crocodilians. *J Zoo Wildl Med* 37:405–408.

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