SPATIAL EFFECTS OF TRANSLOCATION ON GOPHER TORTOISES IN SOUTHEAST GEORGIA

by

LANCE MICHAEL PADEN

(Under the Direction of Kimberly M. Andrews)

ABSTRACT

Modified recreational GPS loggers were used to monitor movements of 18 translocated and 20 resident adult gopher tortoises (*Gopherus polyphemus*) at a southeastern Georgia Wildlife Management Area. Our objective was to determine if home range size differed between sex and residency status. Stationary GPS loggers within tortoise burrows produced a Mean Linear Error of 17.32-72.42m with error corresponding to depth underground. In total, 146,118 successful GPS fixes ($\bar{x}$ = 3,845, range = 493-9,422 per tortoise) were collected in 20 months. There was no difference in resident and translocate home range after penning for 7 and 14 months. Post-penning 95% k-LoCoH mean home ranges were as follows: translocated males= 8.60ha (*n* = 6), translocated females= 2.64ha (*n* = 9), resident males=1.84ha (*n* = 6), resident females= 1.11ha (*n* = 8). Translocates had larger home ranges throughout the cumulative study than residents. Surprisingly, no difference was observed between sexes using GPS data.

INDEX WORDS: Home Range, GPS, *Gopherus polyphemus*, Movement, Resident, Penning, Sex, VHF

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DEDICATION

This thesis is dedicated to my parents, Robert and Nancy Paden. They have always supported and encouraged me in every endeavor in my life and for that I am forever grateful. Some of my earliest memories in life are of being completely captivated by the natural world which they introduced me to and taught me about on countless occasions from a very early age. My father's drive to continually better our family as well as his persistence and professionalism in both his career and many interests have always been traits I have aspired to imitate. My mother has always been there for me with words of encouragement and has always provided me with a constant example of what it means to be a good person and never give up on your passions in life.

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CHAPTER 1

INTRODUCTION AND LITERATURE REVIEW

The Conservation of Gopher Tortoises and Their Habitats

Gopher tortoises (*Gopherus polyphemus*) are terrestrial chelonians endemic to the southeastern Coastal Plain of the United States. In Georgia, they are designated as a state threatened species (Jensen et al. 2008). A unique natural history trait of gopher tortoises is their construction of extensive burrows in sandy soils, thereby providing the services of an ecosystem engineer (Jones et al. 1997). These burrows are used not only by themselves and other gopher tortoise individuals but by a multitude of other vertebrates and invertebrates (Catano and Stout 2015, White and Tuberville 2017). Their extensive burrows act as refugia for many species to survive cool temperatures during winter months and to escape extreme heat and desiccation in the summer months. This trait has made them an iconic ecological "keystone species" throughout their range and, as such, has made their conservation a priority to benefit the many upland commensal species, such as eastern indigo snakes (*Drymarchon couperi*), gopher frogs (*Lithobates capito*), eastern diamondback rattlesnakes (*Crotalus adamanteus*), and Florida pine snakes (*Pituophis melanoleucus mugitus*) that rely on their burrows (Lips 1991, Hyslop et al. 2009, Bauder et al. 2017).

Historically, the Coastal Plain region was home to vast forests of longleaf pine (*Pinus palustris*; Landers et al. 1995) and expansive populations of gopher tortoises associated with this dominant habitat. One of the greatest threats to their survival, as well as many of their commensal species, is that of outright habitat destruction due to anthropogenic development practices (Diemer 1986). Development practices that affect gopher tortoise populations not only include the construction of hard infrastructure such as buildings and roads, but also major landscape alterations such as agricultural, silvicultural, and industrial practices. Interestingly, from 1982 to 2012, croplands in Georgia decreased by 2.6 million acres (U.S. Department of Agriculture 2016). In contrast, pine plantations have become increasingly managed and planted pine stands increased by 4.1 million acres from 1972 to 2014 in Georgia (Brandeis et al. 2014). Following, it is obvious that these agricultural and silvicultural lands comprise a large percentage of land use in Georgia's Coastal Plain and consequently throughout the range of gopher tortoises in Georgia. Some silvicultural practices on industrial timberlands have been shown to increase gopher tortoise burrow abandonment due to canopy closure (Aresco and Guyer 1999, Jones and Dorr 2004). Further fragmentation of remaining suitable habitat occurs due to roads which can be nearly impassable for slow moving tortoises as for many other species (Andrews et al. 2015).

The translocation of gopher tortoises out of areas impacted by development activities occurs throughout their range as a means of avoiding direct mortality (Burke 1989). The process of translocation, moving individuals to an entirely new, yet ecologically suitable area, is gaining ground as an accepted conservation tool (Tuberville et al. 2005, Riedl et al. 2008, Tuberville et al. 2011). Several factors hypothesized to affect the outcome of translocating *Gopherus* spp. populations include: whether tortoises are penned following relocation (Lohoefner and Lohmeier 1986, Tuberville et al. 2005), penning duration (Burke 1989, Tuberville et al. 2005), and prior occupancy of tortoises at the recipient site (Tuberville et al. 2011), translocation distance from the donor site (Hinderle 2011), and habitat suitability at the recipient site (Heise and Epperson 2005, Riedl et al. 2008, Tuberville 2008, Bauder, et al. 2014).

Large home ranges have been documented in the first year post-release, particularly when penning at the recipient site was not performed (Tuberville et al. 2005). Bauder et al. (2014) utilized a 10-month long penning duration and observed high site fidelity among translocated individuals post-release. They suspected that the combination of suitable habitat and penning at the release site contributed to high site fidelity. However, another study compared translocated and resident tortoises' MCP and kernel density home ranges beginning 3 months post-release without any penning at the recipient site and did not observe a statistically significant difference among translocated and resident individuals (Riedl et al. 2008). To ascertain whether a translocation event can be deemed a conservation success, it is important to examine the behaviors and movements of translocated individuals in comparison to resident tortoises at the same site and in the context of understanding the background data on gopher tortoise spatial ecology.

Historic Gopher Tortoise Spatial Ecology

Historically, there have been many spatial use studies conducted on gopher tortoises. The most commonly used methodology for studying spatial use among all *Gopherus spp.* is that of VHF radio-telemetry and the most widely reported metric of home range size has been the Minimum Convex Polygon (MCP). To provide the context of what has been historically documented regarding gopher tortoise space use, Table 1.1 lists the mean and range of Minimum Convex Polygon (MCP) home ranges reported in the literature for gopher tortoises specifically. It is worth noting that many of the studies listed in Table 1.1 had very different main objectives and differed in methodologies so exact comparisons cannot be made without understanding these factors varying among study designs.

There are various intrinsic and demographic factors that have been found to influence home range sizes. One of the most commonly cited conclusions from radio telemetry studies of gopher tortoises is that males have significantly larger MCP home ranges than females (e.g., Guyer et al. 2012, Castellón et al. 2018). See Table 1.1 for a list of such studies (indicated by an asterisk) that have reported statistically significant differences in home range size between males and females. Interestingly, one study in a coastal beach dune habitat of Florida showed no significant difference of sex on MCP home range size although males had statistically significantly larger home ranges when calculated using a fixed kernel method (Lau 2011). Additionally, as tortoises are social creatures that live in colonies, Guyer et al. (2012) reported the importance of density where the greatest home range sizes were observed in colonies existing at a threshold value of 0.4 tortoises/ha, and tortoises occurring at densities both above and below this threshold showed decreased home range sizes. As a final example, tortoise health is another factor which has been shown to have an effect on home range size. Specifically, individuals exhibiting severe symptoms from mycoplasmal upper-respiratory-tract-disease had significantly larger home ranges than mildly symptomatic or asymptomatic individuals (McGuire et al. 2014).

Individual tortoise behaviors and how they interact with their landscapes are also strong drivers of their spatial ecology. Hence, the methods selected for data collection in the field and in the analysis of spatial data are of greatest influence on our understanding of these behaviors. For individuals that exhibit high site fidelity and may only use one or two burrows, MCPs can be a limited home range estimator as creating the polygons require at least three points in space. Somewhat appropriately, some studies either exclude these individuals or assign an extremely small home range value to them such as 0.001 ha. Additionally, MCPs are inherently prone to over-estimating home range size by including areas that are not actually available, much less, used habitat. Therefore, MCPs are limited in allowing for analyses that account for behavioral responses to complex habitat configurations which are especially common in ecologically diverse and human-altered landscapes. Historically in addition to reporting home ranges, VHF radio telemetry studies have also been used to report metrics such as burrow usage and the distance traveled between burrows (McRae et al. 1981, Smith 1995).

Extremely low above-ground observation rates of VHF radio-tracked tortoises in the literature indicate their strong ties to burrows. Eubanks et al. (2003) reported only 0.8% of their 11,423 tracked locations were above ground and 96% of those occurred between the months of June and October. In north-central Florida, a gopher tortoise telemetry study comprised of 5,199 tracked locations resulted in 97 (<2% of total) above-ground observations (Smith 1995). Another study comparing resident and translocated tortoises only observed tortoises on the surface in 0.56% of their 2,320 tracked locations (Riedl et al. 2008). Further, gopher tortoises are often quite cautious when above ground and often will frequently take shelter in their burrows when approached. This is particularly impactful at study sites where telemetered individuals are not acclimated to regular human interactions and immediately seek refuge in their burrows. This tendency for flight upon approach also has presented an increased need to use techniques that minimize disturbance.

The availability of GPS loggers offers the possibility to remotely explore gopher tortoise spatial use in more detail. Secretive behaviors also challenge the viability in using manual radio telemetry to detect non-burrow locations. Hence, there are clear and immediate scientific needs to generate fine-scale datasets (both spatially and temporally) that can allow for comparisons within and among individuals to better understand how gopher tortoises use the landscape when they are not inside their burrows. GPS loggers can obtain location fixes when individuals are outside of their burrows much more frequently than radio tracking alone. This greater degree of spatial use data subsequently allows for more advanced home range analysis techniques, such as the use of estimators like various local convex hull and fixed kernel methods that are more responsive to complexity in tortoise movement behaviors, diverse habitat matrices, and interactions among the two. The availability and miniaturization of GPS loggers paired with advanced analytical home range analysis methods opens opportunities for novel approaches.

GPS Logger Technology and Wildlife Applications

Historically, most of what is known about the spatial ecology of gopher tortoises, like most chelonians, is the result of either VHF radio-telemetry studies or long-term mark-recapture studies (McRae et al. 1981, Kazmaier et al. 2002, Eubanks et al. 2003, Ashton and Burke 2007, Rostal et al. 2014). Manual VHF telemetry allows for certain and direct observation of animals' location and behavior but requires radio tracking individuals frequently and potentially disturbing them or otherwise affecting specific behaviors. These methods certainly offer great benefit to the field of wildlife ecology, but present limitations with some research objectives in that they do not allow for continuous data collection. Continuous data collections allow for greater spatial and temporal resolution that provides a more accurate insight to behavioral patterns and allows for more direct assessment of variation among days, months, seasons, and years. For example, one recent study used commercially available GPS loggers (G30L, Advanced Telemetry Systems Inc., Insanti, MN) and iButtons deployed on Mojave Desert tortoises (*Gopherus agassizii*) to examine how roads and fencing impact their home ranges and behavioral interactions between tortoises and these barriers (Peaden et al. 2017). Such study objectives would not be economical or ethologically sound using manual VHF telemetry alone numerous times a day. As another example, Sperry et al. (2013) utilized radio towers equipped with automated receiving units to remotely acquire continuous spatial data from telemetered ratsnakes (*Pantherophis* spp.) in order to identify seasonal changes in nocturnal activity. While this particular technology allows for continuous data collection, many automated systems are expensive or logistically difficult to install in certain habitats.

Advancements in GPS technology continue to improve fix accuracy, battery life, and decrease weight; however, they are often still prohibitively expensive (Hulbert 2001, Girard et al. 2006) so researchers recently have sought low-cost GPS logger alternatives to gain equivalent ecological insights (Quaglietta et al. 2012, Forin-Wiart et al. 2015). However, Matthews et al. 2013, reported on 24 studies using 280 dedicated wildlife specific GPS collars on terrestrial mammals in Australia. Only 53% of 249 GPS collars actually retrieved/remotely downloaded operated for their full deployment or expected life (with a 101.7 day operating life mean), 40% of studies reported at least one unit failing to collect GPS locations within 10 days of deployment, intermittent GPS collar failures occurred in 31% of deployments, and on average, only 66% of the programmed expected GPS fixes were collected (Matthews et al. 2013). Additionally, the reported cost per fix in Matthews et al. 2013 varied greatly from $0.02 to $129.00 depending on the success of the GPS collars used. Low-cost recreationally marketed GPS loggers are an emerging technology that has been used by researchers to monitor spatial and behavioral activities of cats, dogs, goats, sheep, chimpanzees, turtles, and even humans, as examples (Vazquez-Prokopec et al. 2009, Recio et al. 2011, Forin-Wiart et al. 2015, David Zailo, unpubl. data). Advancements in the affordability GPS technology, such as those that have made i-GotU GT-120 GPS loggers and similarly miniature units available, offer exciting research possibilities that allow researchers to affordably reconsider what is currently known about a species as well as to investigate novel questions.

Impetus and Study Site

In 2015, Southern Ionics Minerals, LLC contracted the Applied Wildlife Conservation Lab (K. Andrews) at the University of Georgia's Odum School of Ecology to develop and manage the environmental stewardship plan for their "Mission Mine." Mission Mine is a heavy mineral sand mine which spans a portion of Charlton and Brantley counties in southeast Georgia. The company primarily mines zirconium and titanium mineral sand deposits on sand ridges which are remnants of barrier islands from Georgia's ancient coastline. These heavy mineral sand deposits are then further refined into Zircon (ZrSiO4), Ilmenite (FeTiO3), and Rutile (TiO2), which are used in a variety of consumer and industrial applications such as high precision aircraft engine casting molds, nuclear fuel rod cladding, white ceramic glazes, paint additives, medical prosthetics, and welding rods. Mission Mine operates on a ridge of Coastal Plain sandhills that predominantly have been in pine silviculture rotation for decades. Southern Ionics Minerals, LLC began mining in Georgia in 2014. As of January 2018, our UGA team has surveyed ~485ha of potentially suitable gopher tortoise upland habitat on potential mining lands, identified 874 gopher tortoise burrows, conducted 277 burrow excavations, nest-searched 235 burrows, and translocated 327 gopher tortoises off site. The company currently employs 140 workers and anticipates mining in the region for the foreseeable future so there is a consistent need for environmental impact mitigation.

A recipient site was necessary for the translocated tortoises. Through collaboration with the Georgia Department of Natural Resources, Penholoway Swamp Wildlife Management Area

(WMA) was selected to receive translocated tortoises in 2016. This WMA was ~63km northeast of Mission Mine in Wayne County, GA and borders the Altamaha River. A ~28ha upland longleaf pine restoration area was chosen on the 1,727ha WMA as our study site due to regular fire management practices and being adjacent to an additional ~150ha longleaf pine restoration area. Additionally, this WMA was determined to have too few resident tortoises to calculate the resident tortoise population density accurately using line-transect-distance sampling protocols (LTDS; Smith et al. 2009). By adding translocated tortoises to this site, there is the potential to augment the population size, facilitating expansion into adjacent restored habitat, and eventually add to the number of minimum viable populations on state-owned lands in Georgia.

Thesis Objectives

First and foremost, I sought to use GPS loggers as a more modern and comprehensive means of assessing the effects of fine-scale behavioral responses of translocated tortoises relative to those of resident tortoises. In particular, were there differences between sexes, residency status (resident or translocated), or an interaction effect of those two factors? Additionally, were there differences (in these factors' effects) present during the penning period and the post penning period?

In order to validate the accuracy of spatial data obtained from our modified recreational GPS loggers on gopher tortoises, these units needed to be tested under the biologically relevant conditions which our study organism experiences. Therefore we sought to determine whether these GPS loggers were capable of obtaining subterranean location fixes and subsequently, we asked, was there a relationship between subterranean depth and Mean Linear Error (MLE) from fixed stationary locations across our study site? This validation process is particularly important for interpreting the accuracy of GPS data from a species that spends much of its life underground.

Table 1.1. Minimum Convex Polygon (MCP) home range sizes reported in the literature for gopher tortoises. An asterisk by the Reference denotes where a study reports significant differences in home range size between males and females. *Mdn* references median rather than mean values.

Male MCP			Female MCP			Study		
n	**Mean (Ha)**	**Range (Ha)**	**n**	**Mean (Ha)**	**Range (Ha)**	**Location**	**Special Notes**	**Reference**
11	28.00	0.40-178.7	9	4.90	0.10-18.30	South-central and east-central GA	10-mo. Pen Transloc-ated	Bauder et al. 2014*
8	3.60	0.1-14.5	7	0.80	0.00-3.50	South-central and east-central GA	Resident	Bauder et al. 2014*
12	6.57	—	11	0.98	—	Central FL	Flatwoods	Castellón et al. 2018*
10	1.24	—	12	3.20	—	Central FL	Scrub	Castellón et al. 2018*
6	0.88	0.00-1.18	5	0.31	0.23-2.88	Central FL	—	Diemer 1992
68	1.10	0.00-4.8	51	0.04	0.00-3.40	Southwest GA	—	Eubanks et al. 2003*
236	.	0.00-15.90	133	.	0.00-8.40	Six sites across range	—	Guyer et al. 2012*
9	0.32	0.13-0.63	11	0.42	<0.01-2.94	Northeast FL	—	Lau 2011
16	1.87	0.11-5.47	14	0.80	0.07-2.54	Southwest GA	95% MCP	McGuire et al. 2014*
8	0.45	0.06-1.44	5	0.08	0.04-0.14	Southwest GA	—	McRae et al. 1981*
3	0.08 (*Mdn*)	—	10	0.34 (*Mdn*)	—	West-central FL	Transloc-ated	Riedl et al. 2008
11	0.63 (*Mdn*)	—	8	0.78 (*Mdn*)	—	West-central FL	Resident	Riedl et al. 2008

6	116.50	0.70-373.70	3	84.20	5.00-145.30	West-central SC	No Pen (Yr 1)	Tuberville et al. 2005*
2	17.50	0.70-34.20	1	5.00	—	West-central SC	No Pen (Yr 1) No dispersers	Tuberville et al. 2005*
5	12.30	0.40-50.20	4	93.90	38.90-134.10	West-central SC	9-mo. Pen (Yr 1)	Tuberville et al. 2005*
4	14.90	0.40-50.20	2	72.20	38.90-105.50	West-central SC	9-mo. Pen (Yr 1) No dispersers	Tuberville et al. 2005*
6	1.40	0.10-5.30	3	4.40	0.10-11.60	West-central SC	12-mo. Pen (Yr 1)	Tuberville et al. 2005
17	23.50	0.20-173.30	10	12.10	0.00-55.00	West-central SC	Yr 2	Tuberville et al. 2005
15	4.40	0.20-15.80	8	2.20	0.00-60.10	West-central SC	Yr 2 No dispersers	Tuberville et al. 2005
11	1.95	0.63-4.89	9	1.07	0.11-2.46	MS	Burned	Yager et al. 2007
9	1.30	0.71-2.43	11	1.90	0.21-7.65	MS	Unburned	Yager et al. 2007
—	—	—	8	0.48	0.00-1.44	Northeast FL	Sandhills	Smith 1995
—	—	—	6	0.11	0.00-0.48	Northeast FL	Old field	Smith 1995
—	—	—	37	0.79	0.01-9.17	Southeast GA	Yr 1	Mitchell 2005
—	—	—	40	1.22	0.01-9.62	Southeast GA	Yr 2	Mitchell 2005

CHAPTER 2
VALIDATION OF MODIFIED GPS LOGGERS FOR USE ON GOPHER TORTOISES

Introduction

Gopher tortoises dig extensive burrows in sandy soils throughout their range in the southeastern Coastal Plain. They rely on their burrows not only for protection from predators and extreme abiotic conditions but also for thermoregulation (Douglass and Layne 1978). Gopher tortoises regularly bask out on their burrow aprons, where they also typically deposit their nests. Although they can be active on the surface when moving between burrows for foraging and mating, we, like many other researchers, have observed via manual VHF radio-telemetry that this species spends much of the time inside their burrows at variable depths. These unique, fossorial natural history traits made stationary testing of our modified i-Got-U GT-120 GPS loggers particularly necessary for the accurate interpretation of spatial data collected. Due to the variability in the quality of spatial data collected among logger types, study sites, and focal species, this site- and equipment-specific validation process has been recommended by other researchers using various GPS loggers (DeCesare et al. 2005, Recio et al. 2011). Further, previous studies have reported increasing linear error among GPS loggers within the same study site due to increasing canopy coverage (Lewis et al. 2007).

We were interested in further testing our GPS loggers in a manner which was biologically catered to the behavior and habits specific to gopher tortoises by deploying them both on the surface and within inactive gopher tortoise burrows across our study site. Specifically, we intended to profile the effect of burrow depth on GPS accuracy at our study site that may be experienced throughout a day in the life of a gopher tortoise. Our objective was to determine the

Mean Linear Error (MLE) of location fixes recorded by GPS loggers at stationary surface points and variable depths inside burrows. This method parallels those that were used in other terrestrial surface stationary testing of GPS units (Yamazaki et al. 2008, Forin-Wiart et al. 2015).

Additionally, we wanted to test a spatial analysis method (nearest-neighbor Local Convex Hulls [k-LoCoH]) on stationary datasets to determine an area estimate of GPS error at three different isopleth percentage values (95%, 75%, and 50%) for the various depths inside burrows or on the surface at our site. These area estimates allowed us to quantify how "noisy" the GPS data are at different depths inside burrows. For biological context, these area estimates also allowed us to approximate the error associated with GPS logger deployed on a gopher tortoise which remained at a single location. Stationary behaviors of tortoises is not uncommon, especially during the cooler months of the year when gopher tortoise activity is minimal.

Finally, several GPS loggers that were deployed on actual tortoises (prior to this stationary testing) randomly shifted time-zones during the deployment which made us question the accuracy of the timestamp data at the hourly temporal scale. Intermittent shifts in duty cycle have been reported to occur in up to 18% of all deployed GPS units (Matthews et al. 2013).

The k-LoCoH home range estimator was selected as an area estimate of GPS error because it only relies on the proximity of location fixes in space rather than a combination of space and time (as with the adaptive LoCoH method, as an example). Erroneous outlier positional fixes obtained with GPS units have the potential to cause Type I error (overestimating the area utilized) in data analysis (Getz and Wilmers 2004). For this reason, a method such as MCPs (which include all of the area contained within the outer-most erroneous fixes) are a less accurate representation of the area in which most of the GPS fixes acquired during deployment are located. Therefore, the k-LoCoH method avoided the use of these intermittently inaccurate time stamps and inherently addressed outlier positional fixes obtained from these GPS loggers based on the designated isopleth percentage value used.

Materials and Methods

GPS Logger Modification

Mobile Action i-GotU GT-120 GPS loggers were modified with larger batteries and waterproofed for this stationary testing study and for use on adult gopher tortoises (D. Zailo, pers comm). The unaltered stock GPS loggers measure approx. 45mm x 22mm x 12mm and weigh 20g with their white plastic housing (Figure 2.1A). To reduce size and to waterproof them, we removed the white plastic housing with a Dremel tool equipped with a Heavy Duty Cut-Off Wheel (Figure 2.1B). This removal was easily achieved by carefully cutting off the end of the GPS logger opposite of the USB cord port. Care was taken to not cut any deeper than the plastic housing itself to avoid damaging the battery or internal hardware. Once the end was off, a small flathead screw driver was used to gently pry off the back panel.

After the back panel was removed, small diagonal cutting pliers were used to snap the remaining housing away from the USB port. Next, WaterWeld® Epoxy Putty, an amount approximately equal in size to that of a pencil eraser, was applied to the back of the USB port to provide reinforcement to prevent it from breaking away. Since these units were only to be deployed for approximately two weeks, we left the stock battery attached (Figure 2.1C, bottom). However, for longer deployments, it is at this point in the modification process that the stock battery could be removed and replaced by soldering on a larger capacity battery for an extended deployment period (see Chapter 3).

The GPS logger battery was charged to full capacity by plugging the USB cord into a power source until charged (typically requiring <24hrs). The loggers then were plugged into a computer with the @Trip PC software installed and the fix schedule was programmed to attempt a fix every 30 min using the Hardware Settings button and following onscreen instructions. Additionally, the indicator lights and button controls were disabled, and the power savings setting was enabled within the Hardware Settings to extend battery life. Once disconnected from the computer, and to ensure the loggers were programmed properly, the power button was held down for three seconds to confirm the button controls were no longer working. Next, the loggers were encased in 1-inch diameter 3M heat shrink tubing and heated using a Wagner 1,200-watt heat gun. The ends of the heat shrink tubing were then crimped using needle-nose pliers and excess tubing trimmed off with diagonal cutting pliers (Figure 2.1C, top).

GPS Logger Deployment and Retrieval

The restoration/relocation area at the Penholoway Swamp WMA was planted in ~5-7yr old longleaf pine with scattered saw palmetto (*Serenoa repens*), turkey oak (*Quercus laevis*), blackberry (*Rubus* spp.), wiregrass (*Aristida stricta*), and eastern prickly pear (*Opuntia humifusa*). In general, the canopy was open throughout the area allowing for sunlight and a plentiful understory that provides forage for tortoises. Five modified GPS loggers were deployed among each of the four stationary location treatment groups: 0.0m (surface), 0.5m inside burrows, 1.0m inside burrows, and 2.0m inside burrows. We randomly selected inactive burrows across the site at which to deploy the GPS loggers. We chose to use currently inactive adult gopher tortoise burrows in order to avoid disturbance of the loggers from tortoises. Additional random locations within the same area were selected for the surface stationary locations. A

12.7mm diameter PVC pipe was cut into 20 segments measuring 30.0cm, 0.5cm, 1.0m, and 2.0m long corresponding to the four location treatment groups (Figure 2.1D). Cable ties then were used to attach the GPS loggers to one end of the pipe. For the loggers deployed subsurface inside of burrows, two 3.175mm holes were drilled through one end of the PVC pipe segment so that the pipe could be held in place from the entrance of the burrow with the square patch antenna (on the GPS logger at the opposite end of the PVC pipe) facing skyward and using 45.72mm long metal tag stakes as anchors. At the surface sites, the 30.0cm PVC pipe segments were driven into the ground ~15cm and the GPS logger attached to the top of the pipe also with the square patch antenna facing skyward. The “true” location of each stationary deployment site was marked using a Garmin GPSmap 64S. It should be noted that as with most comparable handheld GPS units there is also a degree of location error associated with these units used to mark the “true” locations which depends on satellite signal reception. At our study site, it is typical to have a range of 1-4m accuracy when using the Garmin GPSmap 64s.

Sixteen of the GPS loggers were deployed on 17 July 2017, the remaining four GPS loggers were deployed on 19 July 2017, and all were retrieved on 4 August 2017. Once collected, the GPS loggers were downloaded, and the data were trimmed to a uniform start date of 20 July 2017 and a trimmed end date of 2 August 2017. Therefore, the GPS loggers were deployed for a uniform 14 consecutive days, attempting a fix every 30min until either the end of the 14-day period or they ran out of battery.

GPS Logger Data Analysis

We chose to analyze the stationary GPS logger data using two methods. These spatial analyses allowed us to determine both the MLE of these units from the raw data as well as to use a home range estimator in order to calculate an area estimation of the GPS error obtained from stationary locations. To calculate the MLE, analyses were conducted in ArcMap 10.3.1 using the "true" locations (marked with a Garmin GPSmap 64S) and the actual logged data from each stationary GPS logger. Upon importing the Garmin GPSmap 64S and the i-Got-U GT-120 GPS logger data into ArcMap, we projected both datasets into the WGS_1984_UTM_Zone_17N coordinate system. Next, we used the *Point Distance* tool located in the ArcToolBox>Analysis Tools>Proximity folder to calculate the Linear Error (LE) distance to the nearest 0.01m between the "true" logger locations and each of the location fixes acquired by the corresponding GPS loggers during their deployment. The LE distances were recorded in an Excel spreadsheet for all 18 stationary locations that were successful at acquiring satellite fixes during the testing period. The LE dataset was imported into IBM's SPSS statistical software (version 25). A Shapiro-Wilk test of normality was performed on the LE data. Since the data were not normally distributed, a Kruskal-Wallis test (a non-parametric test) was performed to determine if there were any differences between the GPS loggers deployed at different depths. Following, pairwise comparisons were performed on the MLE of the treatment groups using Dunn's (1964) procedure with a Bonferroni correction for multiple comparisons. Adjusted *p*-values were then presented. Additionally, the MLE 95% confidence interval was estimated and graphed.

We chose to use the ZoaTrack.org (formerly oztrack.org) online platform for this analysis because it combines user-friendly features of a Google Earth interface with the flexibility and computational power of adehabitat functions in R through a cloud-based server (Calenge 2006, Dwyer et al. 2015). Another advantage of the ZoaTrack.org platform was the ability to easily export spatial analysis shapefiles (Dwyer et al. 2015). In our case, further post-processing occurred in ArcMap 10.3.1.

We used the ZoaTrack.org online platform to conduct “home range” spatial analyses using the raw data from the stationary GPS logger units. The raw data from each stationary logger was formatted according to the ZoaTrack specifications and uploaded within a single .csv file as a new project. Once the data were added as a new project, each GPS unit’s dataset was selected and three iterations of the nearest neighbor k-Local Convex Hull (k-LoCoH) were performed using 30 nearest neighbors at the 95%, 75%, and 50% isopleths in ZoaTrack using R and adehabitatHR packages on each stationary logger dataset. The *k* parameter value was set to 30 nearest neighbors across each of the 95%, 75%, and 50% isopleth delineations. Each of the resulting isopleths were exported as shapefiles and post-processed in ArcMap 10.3.1. Post-processing in ArcMap required adding an *Area* field to the *Attribute Table* for each of the three k-LoCoH analyses to calculate the area (ha) of each stationary GPS logger’s isopleth. The area (ha) of all of the isopleths was recorded in an Excel table and imported into SPSS for statistical testing.

Results

Mean Linear Error

Although we initially deployed 20 GT-120 GPS loggers as a part of this stationary testing experiment, two of the GPS loggers failed to record locations so only 18 were included in our analyses. The cause of the failures were undetermined; it is possible that they were either damaged during modification or a mistake was made somehow during programming prior to deployment. However, we have found that it is not uncommon to have ~5-10% of these GPS loggers be non-functional even if they are new.

The remaining 18 loggers recorded a total of 7,406 location fixes. Six of these GPS loggers ran out of battery prior to the conclusion of the 14-day deployment. Of those six, all four remaining 2.0m treatment group loggers ran out of battery prematurely as well as one each from the 0.5m and 1.0m burrow depth groups. Figure 2.2 provides examples of the “true” stationary GPS logger locations of four units (one of each burrow depth group) as marked with the Garmin GPSmap 64S handheld unit along with the corresponding recorded modified Mobile Action i-Got-U GT-120 GPS logger fixes.

MLE among the four stationary location treatment groups ranged from 17.32-72.42m, with the surface treatment group having the lowest MLE of 17.32m, and the 2.0m treatment group having the greatest MLE of 72.42m (Table 2.1). The Fix Success Rate (FSR) of attempted location fixes among the four treatment groups ranged from 51% (2m) to 99% (surface; Table 2.1). The median LE for each treatment group was only 20.63-37.83% of that of the MLE, suggesting that the LE dataset contains outliers that are strongly skewing mean estimates (Table 2.1). These LE data also failed the Shapiro Wilk test of normality ($p < 0.05$). A Kruskal-Wallis test was conducted to determine if there were differences in MLE between the different depths: “Surface” ($n = 5$), “0.5 Meter” ($n = 5$), “1.0 Meter” ($n = 4$), and “2.0 Meter” ($n = 4$) inside tortoise burrows. Distributions of MLE values were not similar among groups, as assessed by visual inspection of a boxplot. Differences in MLE between the depths were statistically significant $X^2(3) = 11.075$, $p = 0.011$. *Post hoc* comparisons revealed statistically significant differences in MLE between the surface (4.00) and 2m (15.50; $p = .008$) depths, but not between any other depths. A 95% confidence interval was created for the MLE of each depth (Figure 2.3). In summary, MLE and median LE increased from the surface locations with increasing depth inside gopher tortoise burrows while FSR and total number of fixes decreased with increasing depth inside burrows.

Area Estimate of GPS Error

The estimated area for each of the different depths increased with depth underground for each isopleth percentage. Figure 2.4 displays the isopleths (95%, 75%, and 50%) created from the 18 GPS loggers which were included in the stationary testing analyses. Neither the 75% nor the 50% k-LoCoH isopleths met the normality assumption ($p < 0.05$). Normality was met at the 95% k-LoCoH analysis ($p > 0.05$); however, there were two extreme outliers so a non-parametric test such as the Kruskal-Wallis H test was still an appropriate statistical test for all three analyses. Therefore, three separate Kruskal-Wallis H tests were performed on the area estimate of the GPS error calculated for each stationary logger. Summary statistics for each of those tests are provided in Table 2.2. Both the 95% and 75% k-LoCoH Kruskal-Wallis tests revealed no statistically significant differences in the area estimates of GPS error among the different depths (Table 2.2).

A Kruskal-Wallis test was conducted on the 50% k-LoCoH stationary area estimates of GPS error to determine if there were differences in isopleth area (ha) between the different depths. Distributions of isopleth areas (ha) were not similar for all groups, as assessed by visual inspection of side-by-side boxplots, and were statistically significantly different among depths, $X^2(3) = 10.502$, $p = 0.015$ (Figure 2.5). Subsequently, pairwise comparisons were performed using Dunn’s (1964) procedure with a Bonferroni correction for multiple comparisons to generate adjusted *p*-values. This *post hoc* analysis revealed statistically significant differences in isopleth area (ha) between the surface (mean rank = 4.00) and 2m (mean rank = 15.50); $p = .015$) depths but not between any other depth comparisons.

In summary, Figure 2.5 compares the estimated GPS error isopleths (95%, 75% and 50%) from each stationary location depth. In particular, the 50% k-LoCoH analysis method produced an exceptionally narrow isopleth median range of 0.01-0.09ha (Figure 2.5). However, the 95% k-LoCoH analysis method produced a much broader isopleth median range of 1.56-3.49ha (Figure 2.5), while the 75% k-LoCoH analysis produced an intermediate isopleth median range of 0.06-0.42ha (Figure 2.5).

Table 2.1. Summary data of 14-day stationary testing trial of modified Mobile Action i-GotU GT-120 GPS loggers by depth inside gopher tortoise burrows. Fix Success Rate (FSR) is the rate of successful satellite acquisition of satellite acquisition attempts. Mean Linear Error (MLE) is the mean distance (to the nearest one-hundredth meter) between known "true" locations of stationary loggers as determined by a Garmin GPSmap 64s handheld unit and that of the GT-120 GPS logger fixes. Median Linear Error (*Mdn* LE) is the median distance (to the nearest one-hundredth meter) between the known "true" locations of the stationary loggers and that of the GT-120 GPS logger fixes.

Depth	**N**	**Total Fixes**	**FSR**	**MLE (m)**	***Mdn* LE (m)**	**Std. Dev. (m)**
0.0m (surface)	5	3,172	0.99	17.32	5.51	37.92
0.5m	5	2,199	0.89	26.59	7.56	94.61
1.0m	4	1,369	0.72	37.17	7.67	165.37
2.0m	4	665	0.51	72.42	27.4	218.48

Table 2.2. Kruskal-Wallis H test summary results of stationary testing analyses.

Stationary Analysis	***p*-value**	***χ2(3)***	**0.0m Surface Mean Rank**	**0.5m Mean Rank**	**1.0m Mean Rank**	**2.0m Mean Rank**
95% k-LoCoH	0.355	3.25	7.00	8.60	10.00	13.25
75% k-LoCoH	0.058	7.49	5.50	9.20	9.12	15.25
50% k-LoCoH	0.015	10.50	4.00	9.40	10.50	15.50

Figure 2.1. A. Mobile Action i-GotU GT-120 GPS loggers. B. Using Dremel equipped with Heavy Duty Cut-Off Wheel to remove the plastic housing on GPS logger. C. Modified and numbered stationary GPS loggers (water-proofed with 3M heat-shrink tubing on top, USB connection ports reinforced with WaterWeld at bottom). D. 20 PVC pipe segments (5 each of 30cm, 50cm, 1m, and 2m long) corresponding to 0.0m, 0.5m, 1.0m, and 2.0m inside burrow locations.

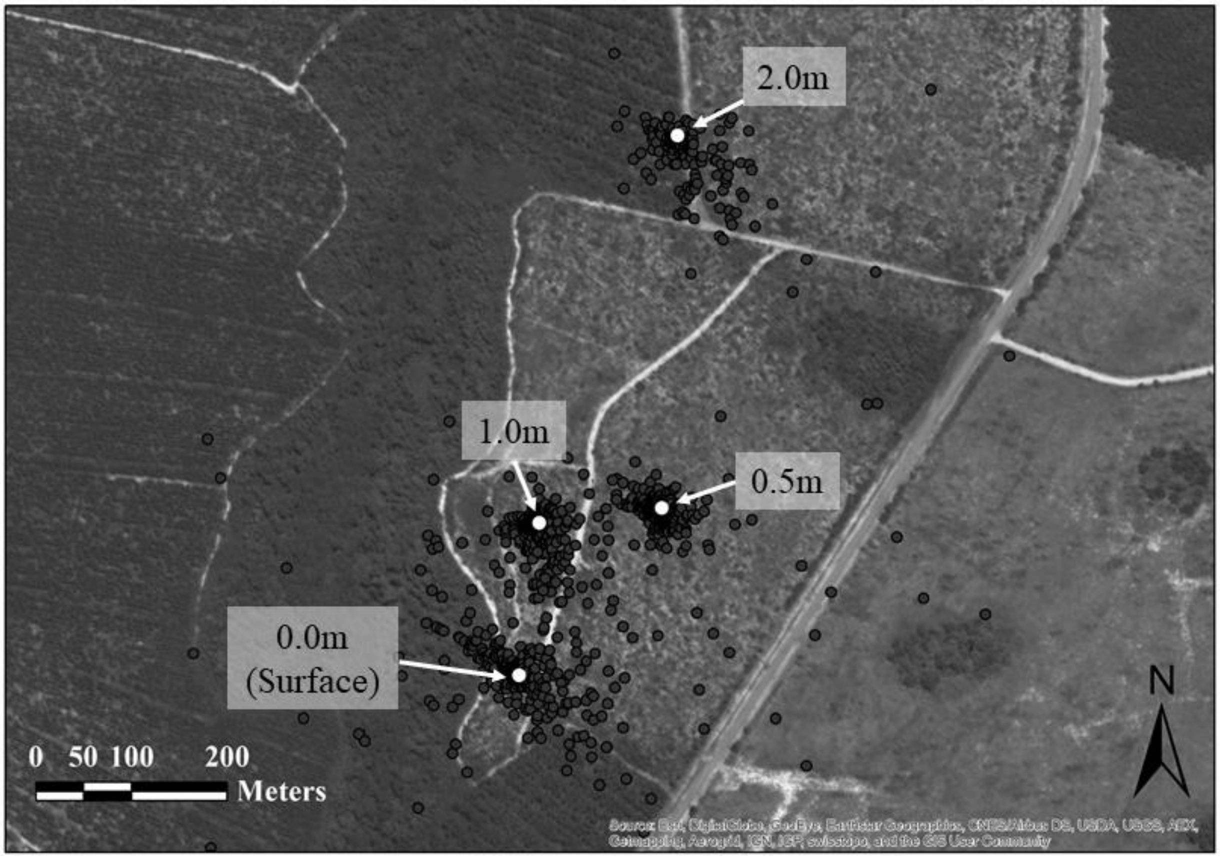

Figure 2.2. An example of the fixes generated from Mobile Action i-Got-U GT-120 GPS loggers deployed at stationary locations at different depths (0.0m, 0.5m, 1.0m, and 2.0m) inside gopher tortoise burrows. The "true" locations of the burrows (as marked with a Garmin GPSmap 64S handheld unit) are shown as a white circle and corresponding GPS logger fixes surrounding each location as gray circles.

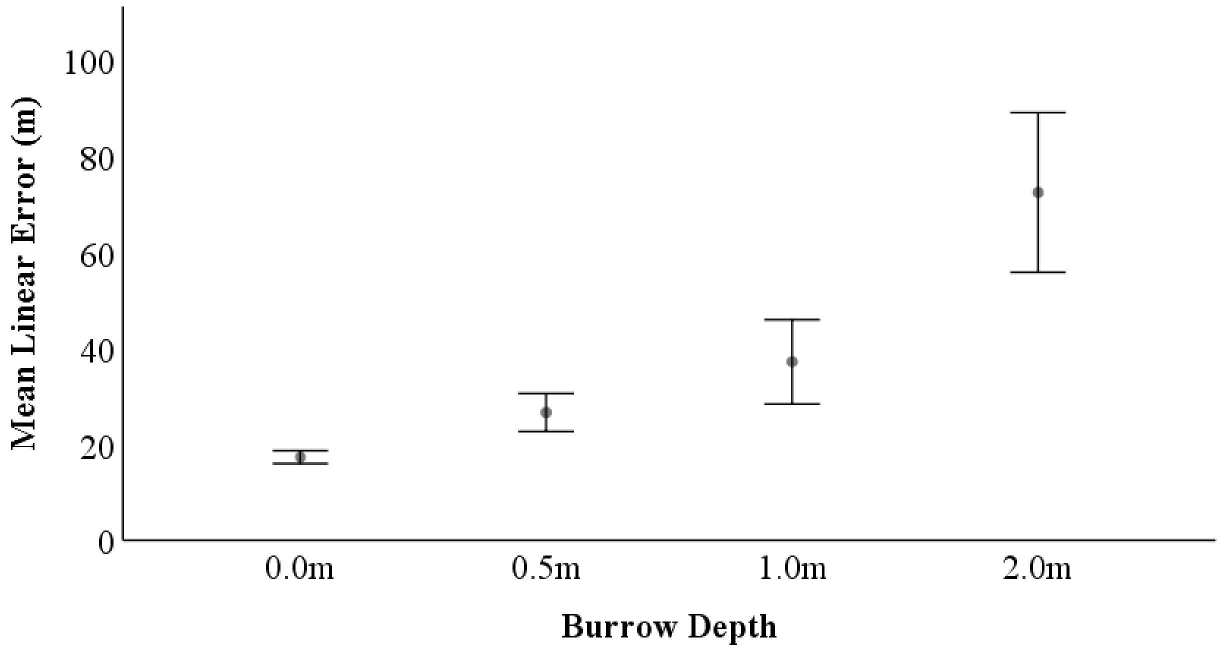

Figure 2.3. Mean Linear Error (m) of stationary GPS logger fixes by depth inside gopher tortoise burrows with 95% confidence intervals. The 0.0m depth corresponded to surface locations of stationary GPS loggers.

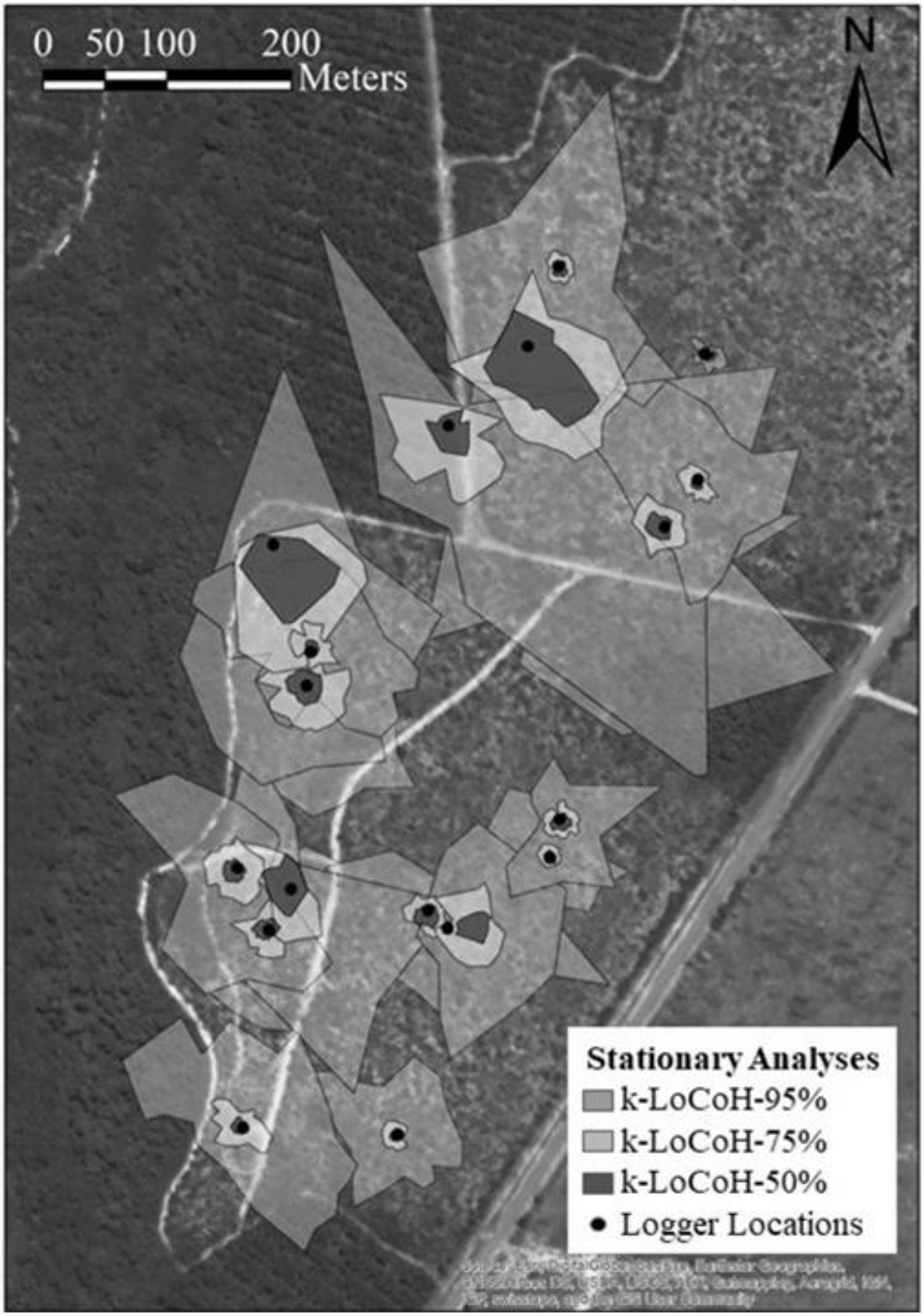

Figure 2.4. Nearest neighbor k-LoCoH isopleths (95%, 75%, and 50%) calculated with the ZoaTrack.org platform corresponding to data from 18 stationary GPS loggers.

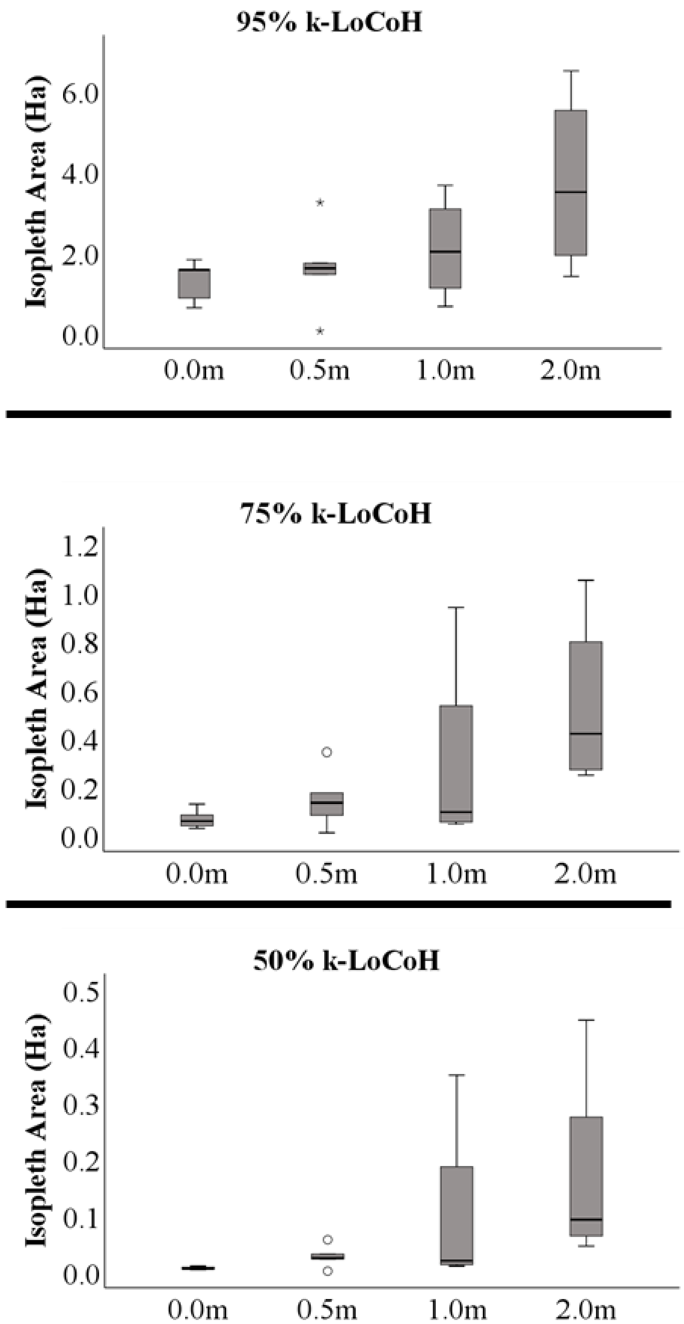

Figure 2.5. Stationary GPS logger spatial analyses using three k-LoCoH isopleth values: 95%, 75%, and 50% (k=30). Isopleth area (ha) is reported for each burrow depth for the respective analyses. The 0.0m depth corresponded to surface locations of stationary GPS loggers. Note: y-axes scale differs among graphs.

CHAPTER 3

GOPHER TORTOISE SPATIAL ECOLOGY: EFFECTS OF TRANSLOCATION

Introduction

The use of ecological mitigation techniques, such as translocating animals (in order to avoid direct mortality) to new presumably suitable habitat, are becoming more commonplace as development increases to match a growing human population. In the case of translocated chelonians, typically larger home ranges are exhibited following their release at the new, recipient site (Hester et al. 2008, Nussear et al. 2012, Farnsworth et al. 2015). Tuberville et al. (2005) demonstrated that temporary penning at the release site was an effective means of increasing site fidelity of gopher tortoises, especially reducing long range movements made within the first year post-release. Additionally, many studies of gopher tortoises have reported that males have significantly larger home ranges than females (indicated by an asterisk, Table 1.1).

As stated in Chapter 1, the objective of this study was to assess the effects of translocation on fine-scale behavioral responses of translocated tortoises relative to resident tortoises. In particular, were there differences between residency status (resident or translocated), sexes, or an interaction effect of those two factors in overall and core home range size? We utilized fine-scale GPS logger data as well as traditional VHF telemetry data in order to compare how translocated and resident tortoises utilized space at our translocation recipient study site. Additionally, since penning has been shown to increase site fidelity of translocated tortoises (Tuberville et al. 2005), it has practical implications for best management practices of translocated gopher tortoises. Therefore, we also used GPS and VHF telemetry data to assess whether there were differences (in sex or residency status on overall and core home range) present during both the penning period and the post penning period? Whereby, the spatial response of translocated tortoises during the post penning period was indicative of acclimation and site fidelity or rather dispersal from the penning location at the recipient site.

Materials and Methods

Donor Site

The Southern Ionics Minerals, LLC Mission Mine site was the donor source of the first group of translocated tortoises used in this study. Burrow surveys and excavations of ~100 gopher tortoise (*Gopherus polyphemus*) burrows were conducted in March 2016 across a ~36ha loblolly pine (*Pinus taeda*) silviculture stand site at Mission Mine that was to be impacted by mining activities later in 2016. An adjacent ~17ha privately owned parcel slated for mining by Southern Ionics Minerals, LLC in 2017 was the donor source of the second group of translocated tortoises used in this study. This upland site predominantly contained a mixture of American turkey oak (*Quercus laevis*), wiregrass (*Aristida stricta*), saw palmetto (*Serenoa repens*), and slash pine (*Pinus elliottii*) as the dominant vegetation. Burrow surveys and excavations of an additional 262 gopher tortoise burrows were conducted in this area from August through October 2016.

Initial Capture and Processing

Burrows were initially identified, flagged, and scoped using a custom burrow scope system (Emmett Blankenship, DVM, Environmental Management Systems) to confirm occupancy. Occupied burrows then were excavated carefully using a PVC pipe guide inside the burrow in order to not lose the track of the burrow during digging (Ashton and Ashton 2008). A mini-excavator operated by a professional were provided by Southern Ionics Minerals, LLC and hand shovels were used to assist with the excavations. Upon excavation, each tortoise ID number was written on the carapace using a Sharpie® until it could be permanently marked. Permanent marks were based on a numeric marginal scute notching system modified from Cagle (1939). The March translocation resulted in a total of 36 tortoises of all age classes that were moved off-site. In September and October 2016, an additional 323 burrows were surveyed resulting in 102 tortoises (hatchlings to adults) to be immediately translocated off-site.

Tortoises were transferred to the Jekyll Island Authority's Georgia Sea Turtle Center (GSTC) in plastic bins where they underwent the following processing procedures. Notch-to-notch carapace length was taken with calipers and used to designate size class as follows: hatchlings: <68mm; juvenile: >68mm & <130mm; subadults: ≥130mm & <230mm (with non-concave plastron); adult males: ≥180mm (with concave plastron); adult females: ≥230mm (non-concave plastron; adapted from Rothermel and Castellón 2014). A determination of sex for adult tortoises was made based on a combination of visual cues of plastral concavity, relative gular length, and carapace length. In general, female gopher tortoises from this site appear slightly larger overall, have shorter gular projections, and minimal plastral concavity in comparison to similar-sized known males.

To avoid injury to the tortoise, while increasing the permanence of the mark, a cordless drill with a preferred 3.175mm drill bit was used to drill the corresponding identification number holes in marginal scutes at approximately 1/3 – 1/2 of the distance from the edge of the marginal scutes for adult and sub-adult tortoises. Large toenail clippers were used to notch a "V" safely into the marginal scutes of juvenile and hatchling tortoises using the same numeric system. The Capture Data Sheet (Appendix A) was an efficient and reliable method for maintaining tortoises' information on capture, excavation, morphometrics, marking, physical exam, laboratory sample checklist, and release. Additionally, health assessments were conducted including oral and cloacal swabs from each tortoise for disease screening as well as blood panels, but results are forthcoming and will not be discussed as a part of this study.

A subset (n = 20) of the translocated adult tortoises were selected for inclusion in our spatial study. From the initial March 2016 translocation period, nine adults (initially all presumed to be male) were selected to receive the initial batch of modified GPS loggers, hereafter referred to as the "Pen 1 group." Therefore, from the second 2016 translocation period, an additional 11 adult female tortoises were selected to receive modified GPS loggers, hereafter referred to as the "Pen 2 group." As a limitation of this study for which we did not intend, comparisons of penning duration are unfortunately confounded by penning duration as well as sex.

Recipient Site

A recipient site was necessary for the translocated tortoises from Mission Mine. Through collaboration with the Georgia Department of Natural Resources, Penholoway Swamp Wildlife Management Area (WMA), was selected to receive translocated tortoises in 2016. This WMA is ~63km northeast of Mission Mine in Wayne County, GA and borders the Altamaha River. An ~28ha upland longleaf pine restoration area was chosen on the 1,727ha WMA as our study site due to regular fire management practices and being adjacent to an additional ~150ha longleaf pine restoration area. In surveys conducted between October 2007 to October 2008, this WMA was determined to have too few resident tortoises to calculate the resident tortoise population density accurately using line-transect-distance sampling protocols (LTDS; Smith et al. 2009). It should be noted that only 6% of 896ha of the suitable upland habitat at the WMA was surveyed during that time. The low densities observed at this site could be attributed to silvicultural practices at the site prior to and at the time of surveys. By adding translocated tortoises to this site, there is the potential to augment the population size, facilitating expansion into adjacent restored habitat, and eventually add to the number of minimum viable populations on state-owned lands in Georgia.

Two adjacent upland pen sites (1.0ha and 2.1ha) at the 1,727ha Penholoway Swamp WMA were identified as suitable locations within which to temporarily contain translocated tortoises (Figure 3.1). The footprint of Pen 1 (1.0ha) was burned several days prior to its installation on 7 March 2016 to increase available new growth forage in this longleaf pine (*Pinus palustris*) restoration area (Figure 3.1). The silt fencing was buried approximately 0.3m deep and starter burrows were constructed using a gas-powered auger for adult burrows and pointed PVC pipes and a mallet for juvenile burrows. Two appropriately sized starter burrows were constructed for each tortoise released into the pen. An attempt was made to distribute the burrows as evenly as possible throughout the pen in an effort to be able to differentiate different adult burrow usage by our selected GPS logger tortoises. The entire Pen 1 group (n = 36 tortoises, n = 9 equipped) was released into the pen on 10 March. Tortoises were monitored closely for signs of stress and over-heating over the next two weeks. One of the study individuals was taken back to the GSTC for a couple days of observed recovery and fluid treatment after it was found flipped on its back and foaming at the mouth. It was confirmed that these symptoms were related to heat stress rather than an infectious disease.

Immediately north of Pen 1 was the site of Pen 2 (2.1ha; Figure 3.1). This area was not burned prior to pen installation; however, it was deemed to have sufficient herbaceous forage for translocated tortoises. On 25 August 2016, Pen 2 (2.1ha) was constructed using heavy equipment and pneumatic tools (Figure 3.1). The Pen 2 group (n =102 tortoises, n = 11 equipped) was added to the pen as they were excavated and processed throughout September and October 2016. Similarly, two appropriately sized starter burrows were created using garden trowels and shovels for each tortoise that was added to the pen. An attempt was made to distribute the burrows as evenly as possible throughout the pen in an effort to be able to differentiate different adult burrow usage by our selected GPS logger tortoises. Formal vegetation surveys were not conducted at either pen site prior to translocation; site judgements were made based on the best available expert opinions through collaboration with Georgia Department of Natural Resources personnel (M. Elliot, J. Jensen, and R. Horan).

Due to the mining schedule for Mission Mine, Pen 1 and Pen 2 were installed at different times in 2016 as described above. However, both Pen 1 and Pen 2 were both deconstructed 8 May 2017, resulting in different penning durations for animals from the two pens (~14 months for pen 1 and ~7 months for Pen 2) but the same start date for the post-penning period. The post-penning monitoring period reported was ~6.5 months. Burrow surveys were conducted at the recipient site prior to pen construction and placement of translocated tortoises into the pens. From these, we know that only a single adult resident and juvenile tortoise resided in the area enclosed by Pen 2; none resided in Pen 1.

In order to establish a comparison of spatial patterns of resident tortoises at the recipient site, we conducted two rounds of trapping to obtain 20 resident tortoises. The first round of trapping took place in “Resident Area 1” from June – August and the second trapping session occurred in “Resident Area 2” from October – November 2016 (Figure 3.1). A total of 20 resident tortoises captured were selected for inclusion in the study as a comparative treatment group with the translocated tortoises. Resident tortoises were always released back into their capture burrow whereas translocated tortoises were placed into either starter burrows or the most recent burrow they were using if re-captured. Resident tortoises were targeted in the surrounding habitat within ~300m of Pens 1 and 2 in order to monitor resident tortoises that would likely come into contact with translocated tortoises once the pens were removed and integration of the populations was possible. Initial capture burrow locations of resident tortoises are indicated by black stars in Figure 3.1.

GPS Logger Modification and Deployment

The i-GotU GT-120 USB GPS loggers (Figure 3.2.A; ~$49.00 each) used on tortoises in this study were modified as outlined in Chapter 2 for the stationary testing. However, equipment weight concerns were reduced given the large size of an adult gopher tortoise and battery life needed to be maximized; hence, the i-GotU GT-120 GPS loggers were modified to use a larger capacity lithium-ion battery similarly to Arnould et al. (2013). The stock units measure ~45mm x 22mm x 12mm with their white plastic housing. To upgrade the mAh capacity of the stock lithium-ion battery and waterproof the GPS loggers, the white plastic housing was removed with a Dremel tool equipped with a heavy duty cut-off wheel (Figure 3.2.B). This modification was easily achieved by carefully cutting off the end of the GPS logger opposite of the USB cord port. Care was taken to not cut any deeper than the plastic housing itself to avoid damaging the battery or hardware within. Once the end was off, a small flathead screw driver was used to gently pry off the back panel. After the back panel was removed, small diagonal cutting pliers were used to snap the remaining housing away from the USB port. As with the stationary loggers, a pencil eraser-sized amount of WaterWeld® Epoxy Putty was applied to the back of the USB port to further support it from breaking away. The stock batteries were then removed by snipping them off with small diagonal cutting pliers. The stock batteries were replaced with either 2200 or 2600mAh lithium-ion 3.7V, 3.5A rechargeable batteries, depending on current availability. A soldering iron was used to solder the aftermarket battery to the GPS logger.

Subsequently, the loggers were plugged into a computer with the @Trip PC software installed and the fix schedule was custom programmed using the Hardware Settings button and following onscreen instructions. GPS loggers deployed in 2017 were programmed to record fixes from 0800-2100 hourly. Initially, in 2016, we attempted a fix every 30min from 0800-2100; however, this fix frequency drained the batteries faster than desired. With hourly fixes from 0800-2100, a 2200-2600mAh lithium-ion battery typically lasts on average ~5.5 months whereas attempting fixes every 30min on the same schedule with the same batteries resulted in ~3 months of battery life. With hourly fixes, we can change batteries twice yearly, once in early spring and again in late fall prior to reduced tortoise activity in the winter. Additionally, indicator lights and button controls were disabled, and the power savings setting was enabled to extend battery life within the Hardware Settings. Once disconnected from the computer, the power button was held down for three seconds to ensure button controls were no longer working and the loggers were programmed properly.

To waterproof the units, the logger batteries were dipped in black Plasti-Dip® nearly to the solder points and allowed to dry for several hours on the shiny (non-stick) side of aluminum foil for additional waterproofing. Once dried, the battery wires were taped down to the GPS using either clear scotch or black electrical tape for added stability and the GPS logger (excluding the battery and battery wires extending beyond the GPS logger) were encased in 2.54cm diameter 3M® heat shrink tubing and heated up using a Wagner 1,200-watt heat gun. The ends of the heat shrink tubing were then crimped using needle-nose pliers while still hot and excess tubing was trimmed off with diagonal cutting pliers.

Once the GPS loggers were successfully programmed and waterproofed, they were attached to the anterior carapace of select adult gopher tortoises. Care had to be taken to arrange the GPS onto the carapace in a way that it did not extend beyond the front of the individual's carapacial marginals as to not impede its ability to enter and exit burrows (Figure 3.3). Importantly, the square antenna of the GPS had to be pointed skyward to ensure and enhance FSR. The VHF radio transmitter was attached to the anterior marginal scutes, and a Thermochron iButton® was attached to one of the anterior most costal scutes as part of a separate study characterizing the thermal profiles (Figure 3.3). WaterWeld® Epoxy Putty was used to affix all equipment to the tortoises' carapaces. The cumulative weight of all devices and WaterWeld epoxy on tortoises was approximately ~175-200 grams. Tortoises ranged in weight from 2.67-7.15kg; therefore, the weight of the equipment relative to the tortoise was typically ~2.8-6.5% of their body weight.

Recaptured tortoises were equipped with a new or re-furbished fully charged and re-programmed GPS logger. Any time the battery was disconnected from the GPS logger, the fix schedule had to be re-programmed prior to re-deployment. Initially, the batteries removed from previously deployed GPS loggers were refurbished by removing the old WaterWeld® and recoating them in Plasti-Dip®; however, this proved to be more time-consuming than was economical so we moved to fully replace the ~$7 batteries with new ones.

As the GPS loggers do not allow us to locate the animals, VHF radio transmitters were also applied to all individuals (Advanced Telemetry Systems, Inc., model: R1680, 3.6g, 189-441 day battery life). Monitoring of both translocated and resident tortoises occurred via VHF radio-tracking on an approximately weekly basis. However, since the primary reason for using the VHF transmitters in this study was to be able to re-capture the tortoises for data downloading and GPS logger swaps, the frequency of manual radio-tracking was not as critical as in dedicated VHF telemetry studies.

GPS Logger Retrieval and Data Downloading

In order to retrieve the data collected by these low-cost GPS loggers, tortoises had to be recaptured and the loggers manually removed for download. Initially, a combination of wire Havahart® traps and bucket traps were deployed at burrows containing telemetered tortoises as determined by the VHF radio-tracking. Bucket traps were placed immediately at the mouth of the burrows and buried flush with the ground (Enge et al. 2012). The buckets were camouflaged using a water-resistant plastic-sided paper drop cloth with sand sprinkled over it (Figure 3.4A). Wire traps were placed at the mouth of tortoise burrows and sunk slightly into the sand so that the wire bottom was not visible and at least half of the trap was shaded with heavy cloth and palmetto fronds. During the hottest months of the year, the traps were checked 3-5 times daily for 4-5 day trapping sessions to avoid the risk of over-heating mortality. This method proved to be highly ineffective. After the initial two ~2 month-long trapping sessions, trapping was restricted to only using bucket traps. Bucket traps were checked twice daily, once around 1000-1100 and again just before sunset (seasonally dependent). If properly shaded with palmetto fronds and available vegetation, the buckets did not overheat to a dangerous temperature.

Additionally, all buckets had 6.35mm holes drilled throughout the bottom of them so that they would drain quickly in the event of heavy rain.

Recaptured individuals were processed at the Georgia Department of Natural Resources Penholoway check station workshop where they received the same suite of capture site and morphometric data collection as was taken at initial capture (Appendix A). The tortoise was placed on a pedestal stand to immobilize the animal without manual restraint and a Dremel equipped with a heavy duty cut-off wheel was used to carefully cut into the WaterWeld® beneath the GPS logger on the upper-side of the GPS logger. The cut had to be ~6-7mm deep underneath the GPS logger and ~1-2mm above the curvature of the carapace on at least one corner to create a good pry point for a flathead screwdriver (Figure 3.4B). A gentle twist and pry motion of a flathead screwdriver allowed the entire GPS logger "tortoise collar" to pop off without harm to the tortoise or the GPS logger and requires minimal effort.

Multiple GPS loggers could be removed and replaced while in the field (or in our case at the workshop) and stockpiled until a later time for downloading the data (Figure 3.4C). Once removed from the tortoise, the end of the GPS logger containing the USB connection port could be carefully revealed using the Dremel and diagonal cutting pliers (Figure 3.4D). The GPS logger is then plugged into a computer to transfer the data using the @Trip PC program. In rare cases, particularly if the battery was completely drained, the GPS logger would have to remain connected to the computer until charged sufficiently to begin the download process. Logger data were saved as a comma-delimited (.csv) file with the individual's identification and download date included in the file name.

GPS Logger Data Analysis

Quality control was performed on all GPS data to remove failed fixes, fixes outside of deployment periods, and obviously erroneous locations (greater than 5km from the study site) and a single master .csv file was created for each individual. The ZoaTrack.org online platform was used to conduct spatial analyses (Dwyer et al. 2015). ZoaTrack was particularly appealing due to its user friendliness as it is a Graphical User Interface program that includes a cloud-based R and Google Earth interface, and it is capable of exporting both .shp and .kml analysis files. The fixed nearest neighbor k-Local Convex Hull (k-LoCoH) analysis method (Getz and Wilmers 2004) was performed in ZoaTrack on each individual using 30 nearest neighbors, and 95% and 50% isopleths were created for home range and core home range estimations, respectively, for each individual. We did not produce 75% isopleths as we did in the stationary trials because we were most interested in overall home range size and highly used areas. The 50% isopleth eliminates all but the most highly used "core" areas within the home range. We chose to use 30 nearest neighbors (after many visual test trials in ZoaTrack) due to the area contained within the resulting 50% isopleth appearing to reach an asymptotic relationship, whereby a greater number of nearest neighbors ('k') did not appear to provide any greater degree of "smoothing" on actual gopher tortoise data, and using fewer than 30 nearest neighbors was prone to inflating 50% isopleths beyond what was considered to be biologically accurate from field observations. In the event that the R script was unable to run one of the analyses at $k = 30$, the next largest k value that would successfully run was used; therefore, we used $k = 30$ as a target parameter value with $k = 30$-38 as the range. This phenomenon appears to be an inherent issue in the R script when using large spatial datasets containing a high degree of clustering.

ZoaTrack performs these analyses through the adehabitat suite of functions (including k-LoCoH) in R (Calenge 2006, Getz et al. 2007). Calculations are performed through R functioning on a cloud-based server and results are displayed onscreen in a Google Earth interface. Each of the resulting isopleths was exported as shapefiles and post-processed in ArcMap 10.3.1. Post-processing in ArcMap required calculating the area contained within the 95% and 50% isopleths, which was achieved by adding an *Area* field to the Attribute Table and using the *Calculate Geometry* option with Precision=10 and Scale=4 in hectares. The area (ha) of all of the 95% and 50% k-LoCoH isopleths was recorded into an Excel table and imported into SPSS for statistical analysis. In order to compare our data to home ranges reported in previous gopher tortoise studies (Table 1.1), we also calculated MCPs using the VHF data for both the cumulative and post penning time periods. We did not calculate MCPs for the GPS data due to the uncertainty associated with filtering out extreme erroneous outlier location fixes which would have greatly artificially inflated an MCP using GPS data from these units. Hence the k-LoCoH method was preferred, which inherently screens for outlier fixes depending on the k-value selected.

The cumulative datasets for each individual were subdivided into penning and post-penning time periods, with 8 May 2017 demarking the start of the post-penning period for both Pen 1 and Pen 2 tortoises. Both 95% and 50% k-LoCoH analyses were conducted in ZoaTrack on each individual for each period following the procedures described previously. Shapiro-Wilk tests of normality were conducted, and bar plots were used to identify outlier data. Because these data were not normally distributed, nor could they be transformed to meet the normality assumption while retaining biologically relevant outlier data, a non-parametric test was necessary. An advantage to employing weekly VHF radio tracking was that it confirmed that some of the furthest points were true tortoise locations. For analyses testing the effect of residency status, translocated individuals were pooled together from both Pen 1 and Pen 2. Subsequently, Mann-Whitney U tests were performed individually for factors of sex and residency status at the 95% k-LoCoH, 50% k-LoCoH, and 100% MCP home ranges for each of the study periods (cumulative study period, penning period only, post-penning period only). Furthermore, a Kruskal-Wallis (non-parametric) test was performed to test for an interaction effect of sex and residency status for any time period which had individually statistically significant effects of either sex or residency status.

Results

Cumulative Study Period

Of the 40 individuals equipped with GPS loggers, two GPS loggers (deployed on female translocated tortoises) failed to record any GPS data on multiple deployments and were therefore excluded from the GPS analyses. A total of 146,118 successful GPS fixes ($\bar{x}$ = 3,845, range = 493-9,422) were collected between 10 March 2016 and 23 November 2017. The number of successful GPS fixes as well as VHF tracks by individual are included in Appendix B. Mean 95% k-LoCoH home ranges in order of decreasing area were: translocated males = 4.47ha (*n* = 6), translocated females = 4.08ha (*n* = 12), resident males = 2.58ha (*n* = 9), resident females = 2.08ha (*n* = 11; Table 3.1). Mean 50% k-LoCoH core home range values in order of decreasing area were: translocated males = 0.10ha (*n* = 6), translocated females = 0.07ha (*n* = 12), resident males = 0.06ha (*n* = 9), resident females = 0.04ha (*n* = 11; Table 3.1). Mean 100% MCP home range values in order of decreasing area were: translocated males =17.08ha (*n* = 6), translocated females = 4.46ha (*n* = 12), resident males = 4.37ha (*n* = 9), resident females = 0.94 (*n* = 9; Table

3.1) For comparison to home ranges from other gopher tortoise studies (Table 1.1), MCPs from manual VHF tracking are also reported by penning treatment for individuals throughout the cumulative and post penning study periods which were radio-tracked to at least three unique locations during the study (Table 3.2).

Interestingly, no statistically significant difference was observed via Mann-Whitney U tests among sexes during this study at either the 95% k-LoCoH home range or the 50% k-LoCoH core home ranges (Table 3.3). Figure 3.5 illustrates this unexpected result in the form of side-by-side boxplots of males (n = 15) and females (n = 23). Likewise, no significant difference among sexes was observed using 100% MCPs from VHF telemetry data (U = 139, z = -1.000, p = 0.329; Table 3.3).

Two outlier individuals were identified from bar plots of residency status and 95% and 50% k-LoCoH home ranges. In Mann-Whitney U tests for assessing the influence of residency status, distributions of both the home range and core home range areas for resident and translocated groups were not similar, as assessed by visual inspection of population pyramid SPSS output (Figure 3.6). Comparisons of mean ranks are reported instead of group medians due the non-similar distributions observed. The 95% k-LoCoH home ranges for the translocated group (mean rank = 24.06) was significantly greater than for the resident group (mean rank = 15.40, U = 98, z = -2.397, p = 0.016; Table 3.3). Likewise, core home ranges for the translocated group (mean rank = 23.58) were also statistically significantly greater than for the resident group (mean rank = 15.82, U = 106.5, z = -2.149, p = .030; Table 3.3). A statistically significant difference between residency status was also observed using 100% MCPs from VHF telemetry data (U = 102, z = -2.280, p = 0.022; Table 3.3). Although we tested for an interaction effect of sex and residency on home range size using a Kruskal-Wallis H test, there were no significant differences among the above sex and residency groups (Figure 3.7).

Penning Period

Of the 40 individuals initially equipped with GPS loggers, 28 were included in the penning period analyses (Appendix B). The included individuals were ones for which we had GPS data available during both the penning and the post penning period. A total of 85,033 successful GPS fixes ($\bar{x}$ = 3,037, range = 1,049-7,324) were collected between 10 March 2016 and 23 November 2017. Shapiro-Wilk tests of normality were conducted on the 95% and 50% k-LoCoH home ranges by sex and residency status and were shown to be not normally distributed ($p < 0.05$). Mann-Whitney U tests were conducted on penning period data to determine if there were differences in 95% k-LoCoH home range and 50% k-LoCoH core home range size between sex and residency status. This analysis includes pooled residency status (Pen 1 and Pen 2 translocates vs. residents) data from individuals from the start of the study until 8 May 2017 when deconstruction of the pens occurred. No statistically significantly difference was observed among sexes at the 95% k-LoCoH home ranges ($p > 0.05$) nor the 50% k-LoCoH core home ranges ($p > 0.05$) specific to the penning period (Table 3.3). Interestingly, no statistically significant difference was observed among residency status at the 95% k-LoCoH home range ($p > 0.05$). However, there was a statistically significant difference between residency status at the 50% k-LoCoH core home range ($p < 0.05$; Table 3.3).

Post-Penning Period

The same 28 individuals (for which we had GPS data available during both the penning period as well as the post penning period) were included in the post penning period analyses as during the penning period (Table 3.1). This time period includes pooled data from individuals from 8 May 2017 (when deconstruction of the pens occurred) until 23 November 2017. A total of 38,861 successful GPS fixes ($\bar{x}$ = 1,388, range = 298-2,326) were collected between 8 May 2017 and 23 November 2017. Shapiro-Wilk tests of normality were conducted on the 95% and 50% k-LoCoH home ranges by sex and residency status and were shown to be not normally distributed ($p < 0.05$). Mann-Whitney U tests were conducted on post penning period data to determine if there were differences in 95% k-LoCoH home range and 50% k-LoCoH core home range size between sexes. No statistically significant difference was observed among sexes during this study at the 95% k-LoCoH home range nor the 50% k-LoCoH core home ranges during the post-penning period (Table 3.3). However, we did see a difference among sexes when using MCPs from VHF telemetry data ($p < 0.05$; Table 3.3). Mean 100% MCP post-penning home range values in order of decreasing area were: translocated males =17.00ha ($n = 5$), resident males = 2.90ha ($n = 9$), translocated females = 1.37ha ($n = 12$), resident females = 0.74 ($n = 9$; Table 3.1). No statistically significant differences were observed between translocates and residents during the post-penning period ($p > 0.05$; Table 3.3).

Table 3.1. Summary of cumulative (10 March 2016 to 23 November 2017) and post-penning (8 May 2017 to 23 November 2017) home ranges (ha) by sex and residency status. Note that *n* varies between the cumulative and post penning time periods as well as between GPS and VHF data based on the ability to calculate an MCP by having three or more unique location fixes for an individual.

Residency Status	**Time**	**Sex**	***n***	**95% k-LoCoH Mean (Range)**	**50% k-LoCoH Mean (Range)**	***n***	**MCP Mean (Range)**
Resident	Cum.	♂	9	2.58 (0.59-4.80)	0.06 (0.02-0.13)	9	4.37 (0.21-22.46)
	Cum.	♀	11	2.08 (0.71-3.83)	0.04 (0.01-0.12)	9	0.94 (0.06-1.85)
Translocated	Cum.	♂	6	4.47 (1.06-13.98)	0.10 (0.05-0.22)	6	17.08 (0.66-66.60)
	Cum.	♀	12	4.08 (2.04-12.33)	0.07 (0.03-0.11)	12	4.46 (0.34-12.29)
Resident	Post	♂	6	1.84 (0.26-4.85)	0.04 (0.01-0.07)	9	2.90 (0.18-15.34)
	Post	♀	8	1.11 (0.21-2.22)	0.03 (0.01-0.06)	9	0.74 (0.06-1.62)
Translocated	Post	♂	5	8.60 (0.26-30.83)	0.08 (0.01-0.25)	5	17.00 (0.14-60.71)
	Post	♀	9	2.64 (0.79-6.77)	0.03 (0.01-0.05)	12	1.37 (0.05-5.06)

Table 3.2 Post-penning Minimum Convex Polygon (MCP) home ranges reported from ours and similar translocated and resident tortoise studies using VHF radio telemetry data. *Mdn* references median rather than mean values. Table adapted from Bauder et al. (2014, Table 2).

Study					**Male MCP (Ha)**		**Female MCP (Ha)**	
Reference	**Location**	**Treatment**	**Starter burrows?**	**Post-release monitoring duration**	***n***	**Mean (Range)**	***n***	**Mean (Range)**
This study	Southeast GA	7-mo. Pen	Yes	~6.5 mo.		NA	10	1.49 (0.05-5.06)
This study	Southeast GA	14-mo. Pen	Yes		5	17.00 (0.14-60.56)	2	0.77 (0.24-1.30)
This study	Southeast GA	Resident			9	2.90 (0.18-15.34)	9	0.74 (0.06-1.62)
Bauder et al. 2014	South-central and east-central GA	10-mo. Pen	Yes	5 mo.	11	28.00 (0.40-178.7)	9	4.90 (0.10-18.30)
Bauder et al. 2014	South-central and east-central GA	Resident			8	3.60 (0.1-14.5)	7	0.80 (0.00-3.50)
Riedl et al. 2008	West-central FL	No Pen	No	15 mo.	3	0.08 (*Mdn*)	10	0.34 (*Mdn*)
Riedl et al. 2008	West-central FL	Resident			11	0.63 (*Mdn*)	8	0.78 (*Mdn*)
Tuberville et al. 2005	West-central SC	No Pen	Yes	2-5 mo.	6	116.50 (0.70-373.70)	3	84.20 (5.00-145.30)
Tuberville et al. 2005	West-central SC	9-mo. Pen	Yes		5	12.30 (0.40-50.20)	4	93.90 (38.90-134.10)
Tuberville et al. 2005	West-central SC	12-mo. Pen	Yes		6	1.40 (0.10-5.30)	3	4.40 (0.10-11.60)

Table 3.3. Test results for Mann-Whitney U tests for factors of sex and residency status at the 95% and 50% k-LoCoH home ranges (HR) for each of the study periods using GPS logger data: cumulative study period (Cum.; *n* = 38), penning period only (Pen; *n* = 28), and post-penning period only (Post; *n* = 28). Cumulative MCPs (100%) calculated from VHF telemetry data (*n* = 38). Post-penning period MCPs (100%) calculated from VHF telemetry data (*n* = 35). Mean rank values are reported instead of *Mdn* HR for tests which had non-similar distributions (as required by the assumption of similar distributions necessary for Mann-Whitney U tests).

Variable	**HR**	**Time**	***p*-value**	***U***	***z* score**	**♂ *Mdn* HR (ha)**	**♀ *Mdn* HR (ha)**	**♂ Mean Rank**	**♀ Mean Rank**	**Resid. Mean Rank**	**Trans. Mean Rank**
Sex	95%	Cum.	0.86	179.0	0.194	2.43	2.72	—	—	—	—
	50%	Cum.	0.202	129.0	-1.299	0.06	0.05	—	—	—	—
	MCP	Cum.	0.329	139.0	-1.000	—	—	21.73	18.04	—	—
Residency	**95%**	**Cum.**	**0.016**	98.0	-2.397	—	—	—	—	15.4	24.06
	50%	**Cum.**	**0.03**	106.5	-2.149	—	—	—	—	15.82	23.58
	MCP	**Cum.**	**0.022**	102.0	-2.280	—	—	—	—	15.17	23.4
Sex	95%	Pen	0.051	238.0	1.956	1.71	2.38	—	—	—	—
	50%	Pen	0.202	129.0	-1.299	—	—	22.4	17.61	—	—
Residency	95%	Pen	0.196	135.0	-1.316	—	—	—	—	17.25	22
	50%	**Pen**	**0.033**	107.5	-2.120	—	—	—	—	15.88	23.53
Sex	95%	Post	0.817	88.0	-0.259	1.57	1.24	—	—	—	—
	50%	Post	0.378	74.5	-0.894	—	—	16.23	13.38	—	—
	MCP	**Post**	**0.024**	80.0	-2.256	—	—	22.79	14.81	—	—
Residency	95%	Post	0.178	68.0	-1.378	—	—	—	—	12.36	16.64
	50%	Post	0.946	96.5	-0.069	—	—	—	—	14.39	14.61
	MCP	Post	0.503	132.0	-0.693	1.47	0.7	—	—	—	—

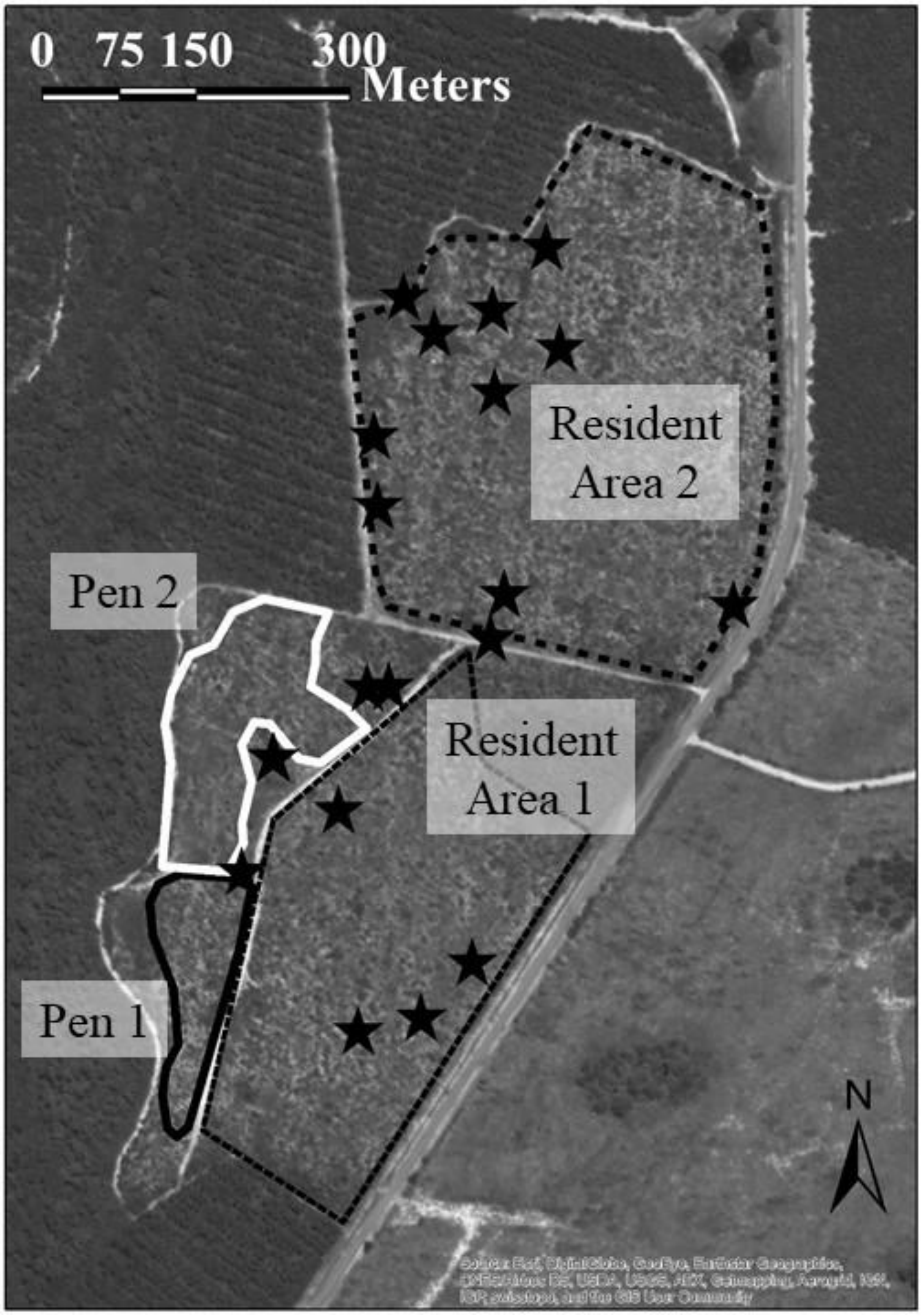

Fig. 3.1. Arrangement of Pen 1 (1.0ha, in solid black) and Pen 2 (2.1ha, in solid white) at the Penholoway Swamp WMA study site. Black stars indicate initial capture burrow locations of resident tortoises. Resident Area 1 (and the area immediately east of Pen 2) were the sites of resident tortoise trapping efforts from May-July 2016. Resident Area 2 was the site of additional resident tortoise trapping in October-November 2017.

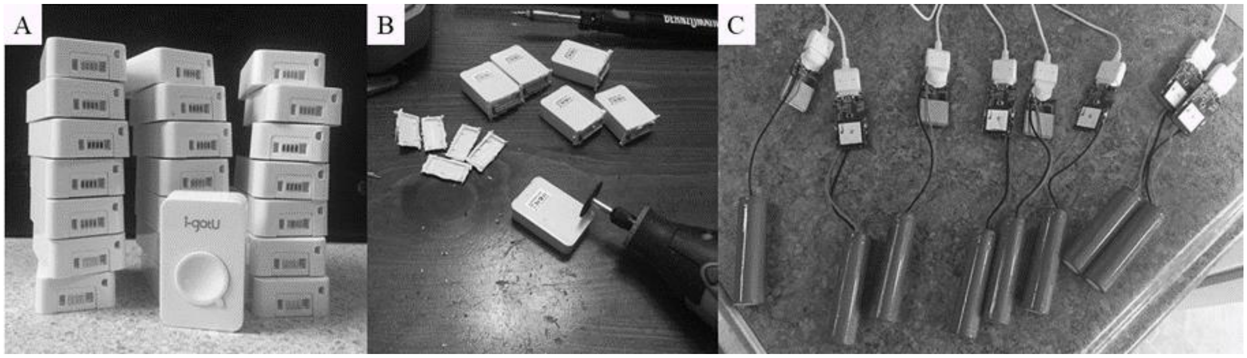

Figure 3.2. A. i-GotU GT-120 USB GPS Loggers. B. Using a Dremel with cut-off wheel to remove plastic housing. C. GPS loggers charging the newly soldered lithium-ion rechargeable batteries

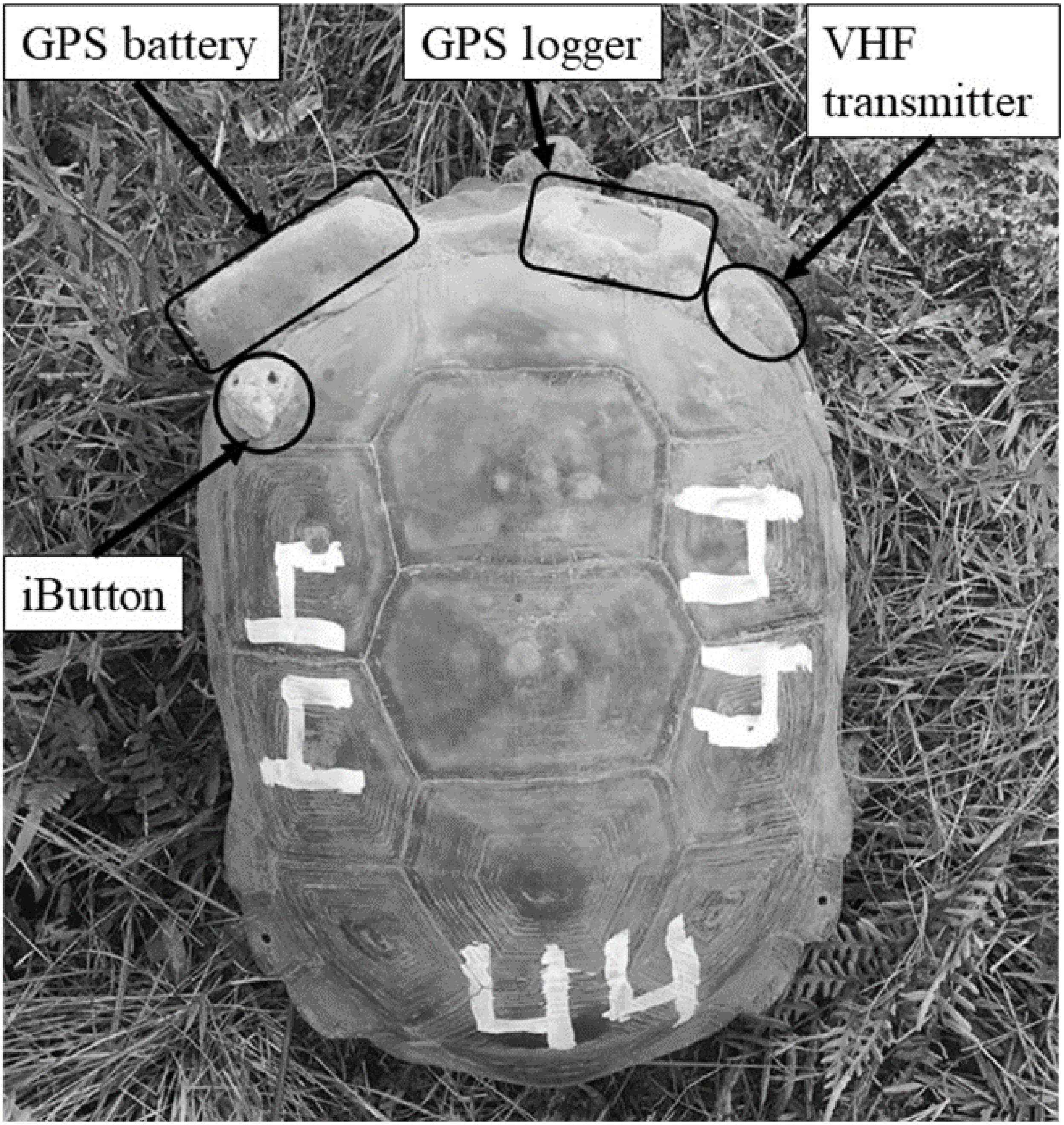

Figure 3.3. Example attachment diagram for the all of the equipment attached to our study individuals (GPS logger, GPS battery, VHF radio transmitter, and Thermochron iButton®).

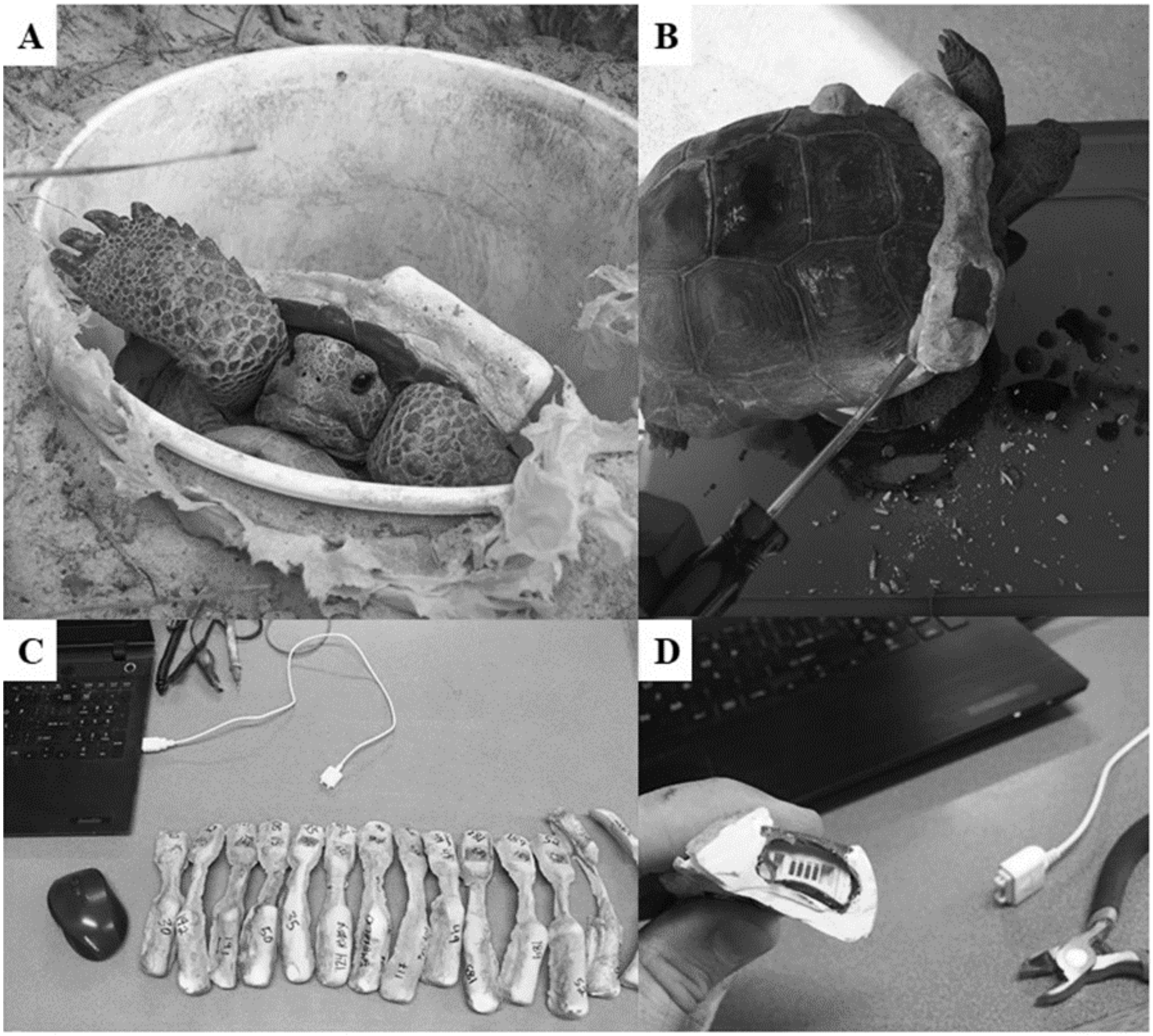

Figure 3.4. A. Recapture of a tortoise using a bucket trap. B. GPS logger removal using a flathead screwdriver after creating a pry-point with a Dremel. C. Retrieved GPS loggers. D. Removed end for USB download.

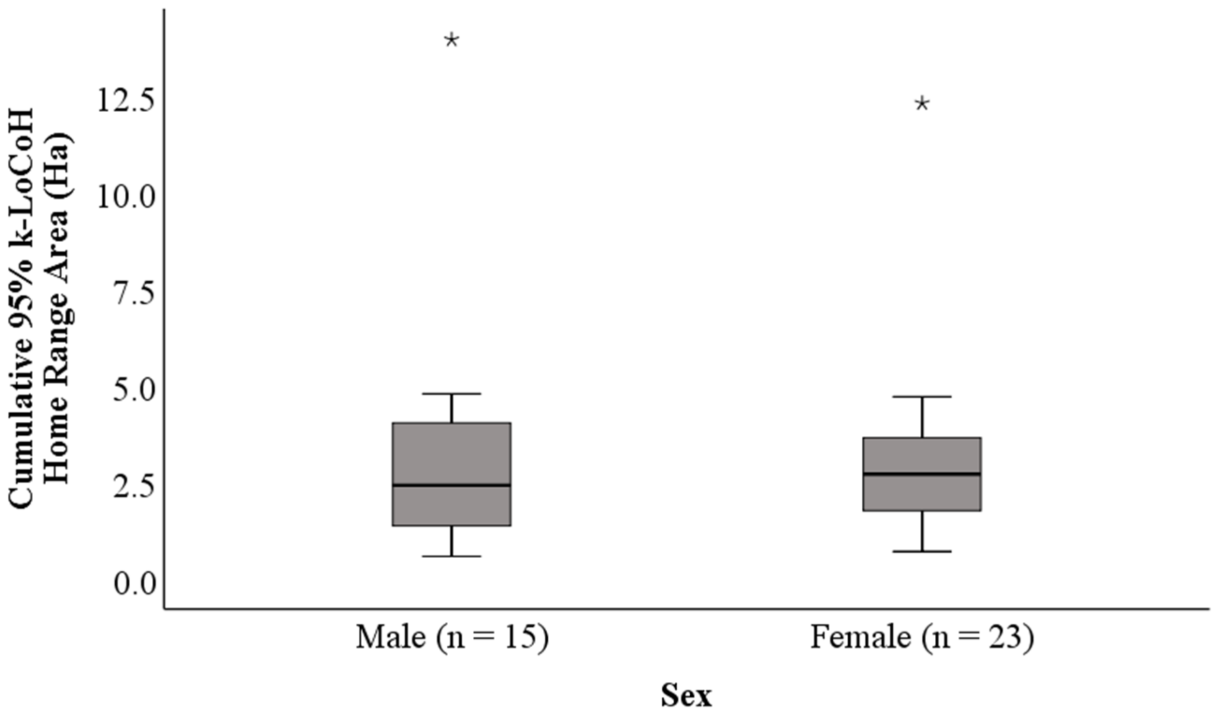

Figure 3.5. Gopher tortoise home range area (ha) by sex from GPS logger data. Translocated and resident individuals were pooled (n = 38) throughout the study to test for an effect of sex on home range size. This cumulative study period ranged from 10 March 2016 to 23 November 2017. The 95% k-LoCoH home ranges were constructed from 146,118 successful GPS fixes ($\bar{x}$ = 3,845, range = 493-9,422).

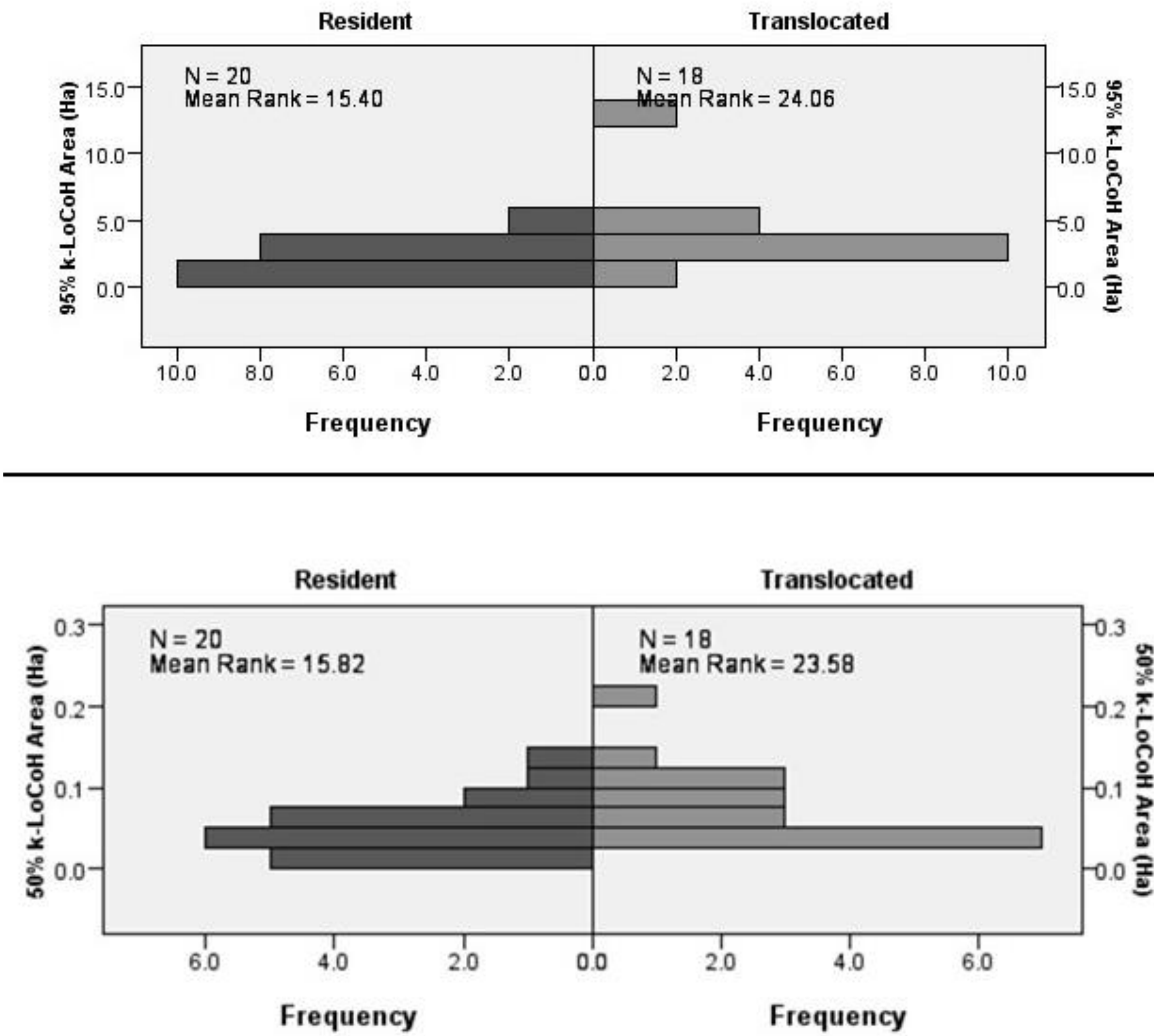

Figure 3.6. Mann-Whitney U test output from SPSS (version 25) showing non-similar distributions of home ranges of gopher tortoises by residency status for 95% (top) and 50% (bottom) k-LoCoH home range sizes.

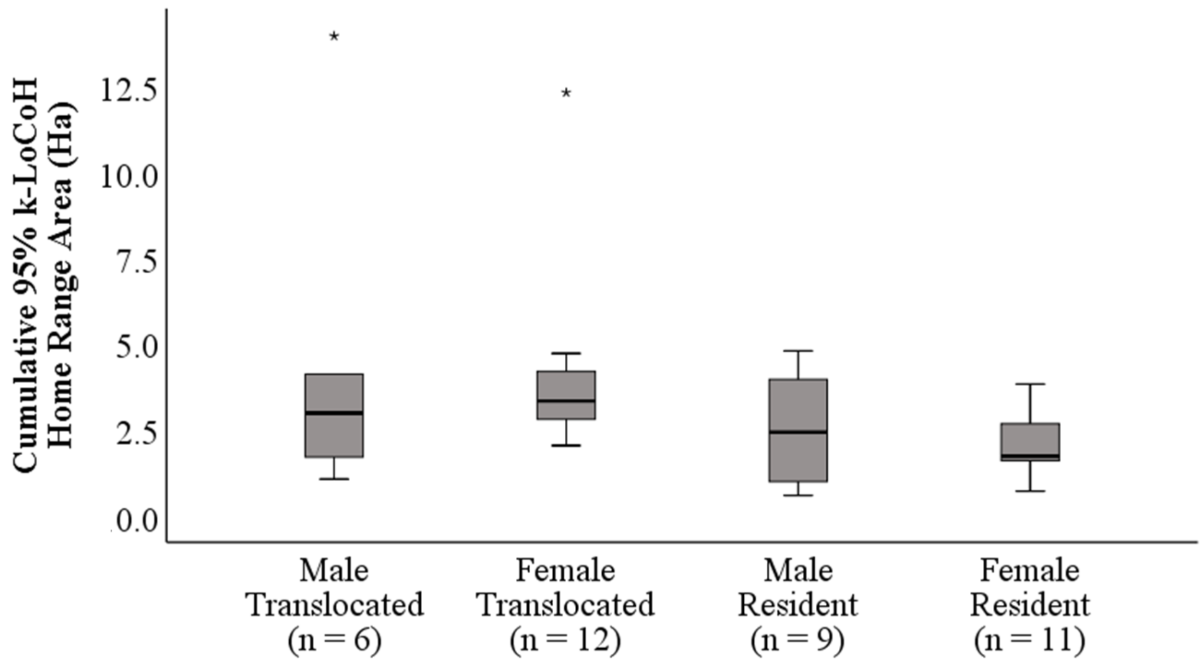

Figure 3.7. Gopher tortoise home range area (ha) by sex and residency status from GPS logger data. This cumulative study period ranged from 10 March 2016 to 23 November 2017. The 95% k-LoCoH home ranges were constructed from 146,118 successful GPS fixes ($\overline{x}$ = 3,845, range = 493-9,422).

CHAPTER 4

DISCUSSION

As anthropogenic activities continue to impact gopher tortoise populations, the need for environmental impact mitigation techniques such translocations will remain. By monitoring the spatial movements of both translocated and resident tortoises at a protected recipient site, we were able to examine how translocation affects the space use of both groups. Surprisingly, we observed no statistically significant difference in space use at the site between resident and translocated tortoises over ~5.5 months of monitoring following two temporary penning treatments of 7 to 14 months. Analyses included two home range estimates (a 95% k-LoCoH analysis for GPS logger data and the historically relevant 100% MCP analysis for manual VHF telemetry data) as well as a core home range estimate (50% k-LoCoH analysis for GPS logger data). This finding was surprising because it is well documented in the literature that translocated chelonians exhibit larger home-ranges post-release than resident individuals (Hester et al. 2008, Nussear et al. 2012, Farnsworth et al. 2015). Our data do follow the generally reported trend of translocated tortoises having larger home ranges than residents despite the lack of statistically significant differences between them. Our post-penning MCPs appear to fall within the range of values reported by similar translocate-resident gopher tortoise studies (Table 3.2). In particular, our resident male (2.90ha) and resident female (0.74ha) home ranges are very similar to those of Bauder et al. (2014) who reported home range use of resident males (3.60ha) and resident females (0.80ha). However, our study observed smaller home ranges of translocated individuals with males using a mean of 17.00ha and females using a mean of 0.77-1.49ha (depending on penning duration). Our translocated males and females also had smaller home ranges than those reported by Tuberville et al. (2005) who reported treatment means of males ranging from 1.40-116.5ha and females ranging from 4.40- 93.90ha. Perhaps our study may not have observed any statistically significant difference in home range size between resident and translocated tortoises during the post-penning period as a result of increased site fidelity due to temporary penning prior to release (Tuberville et al. 2005). The larger post-penning home ranges that we report in comparison to Riedl et al. (2008) might be attributable to including the first three months of data post-release in our analyses.. Available habitat of a relatively high quality due to prescribed fire management and longleaf restoration at the site could also have contributed to translocated individuals remaining at the site following release. Also, tortoise density at the site could have potentially affected the space use patterns we observed since the tortoise density is clearly (although not calculated specifically) greater than the maximum home range threshold density of 0.40tortoises/ha reported (Guyer et al. 2012). Another recent study using a large VHF telemetry dataset from resident and translocated Herman's tortoises (*Testudo hermanii hermanii*) demonstrated findings of relatively rapid settlement of translocated individuals (Pille et al. 2018).

The other surprising result we observed from GPS data was a lack of statistically significant difference between home range size of males and female gopher tortoises from our GPS data. This finding is somewhat unusual in gopher tortoise studies because there have been so many historical studies which have reported significant differences between sexes (McRae et al. 1981, Eubanks et al. 2003, Tuberville et al. 2005, Guyer et al. 2012, Bauder et al. 2014, Castellón et al. 2018). However, we did observe a significant difference in sex from our MCPs created from VHF telemetry data. This discrepancy illustrates how different our perceptions of space use can be when using various analysis methods. While MCPs are sufficient for identifying home ranges for species across homogenous suitable habitat, they perform poorly when lengthy movements are made across non-suitable habitat. In such scenarios, MCPs are prone to greatly overestimating the area an individual utilizes. While no statistically significant differences between sexes were detected for home range size from GPS logger data, this non-significance is not necessarily an insignificance as it does not mean that male and female tortoises are using the landscape in the same way. Perhaps males make more regular short-term movements between burrows in their home ranges than females who may be more prone to spending more time at each burrow they utilize in their home range. This hypothesis is anecdotally consistent with our observations from VHF radio tracking. However, these short-term movements are difficult to distinguish from linear errors of GPS loggers at nearby locations when data are pooled over longer periods. Given the understandings of both tortoise spatial ecology and tracking methods, we could be in a better position to pursue this as an interesting future question using GPS loggers.

In review of our modified GPS loggers, these units performed reasonably well when deployed on gopher tortoises. However, on average, several GPS loggers failed between each data downloading session (every 3-6mo). Likewise, we experienced two GPS logger failures during stationary testing. We estimated a failure rate of ~5-10% during each deployment period (3-6 mo). While not definitively determined, these failures may have been caused by issues in the manufacturing of these low-cost units or during our modification process. Some of these failures could also be attributed to accidentally damaging them during removal from the carapace. This type of failure may be further reduced by using a more temporary attachment method similar to the brackets used by Peaden et al. (2017).

Matthews et al. (2013) compiled a list of common problems from 24 GPS studies representing 280 GPS collar deployments on terrestrial mammals in Australia. Some of those common problems reported and their pervasiveness which are relevant to our study included: shorter GPS life than expected (affecting 47% of GPS collars), intermittent fixes (affecting 31% of GPS collars), fewer fixes per day than expected, changes to fix schedule or duty cycle (affecting 18% of GPS collars), VHF transmitter failure (affecting 14% of GPS collars) (Matthews et al. 2013). In addition to the aforementioned GPS logger failures, we also experienced shorter GPS life than expected, particularly during the first deployment of nine GPS loggers on the Pen 1 group. This occurrence was remedied by adjusting the fix interval to 60 minutes to increase GPS deployment life. We also encountered several instances of VHF transmitter failure throughout our study in the form of broken antennas and premature transmitter exhaustion which lead to temporarily missing individuals which sometimes affected how quickly we were able to re-capture those individuals to download and replace their GPS logger and VHF transmitter. All of the aforementioned sources of failure can lead to lost GPS data and attempts should be made to minimize these problems. Issues such as these should be factored in accordingly from the onset of study design by anticipating failure rates based this and other studies.

Beyond outright GPS logger failure, we observed decreased Fix Success Rate (FSR) with increasing depth inside burrows. While in general, gopher tortoises are a highly subterranean species, individual variation in surfacing behaviors and burrow depth can result in fewer GPS fixes than expected. For example, a tortoise extensively using a particularly deep burrow will result in a low FSR thereby limiting the number of overall location fixes collected for that individual. GPS fix-rate bias can be a concern for GPS studies because it can potentially lead to inaccurate interpretations of spatial use due to lost data under particular environmental conditions (D’Eon 2003). Other studies using GPS loggers have examined how factors such as canopy cover, elevation, and movement impact both the MLE and FSR of GPS loggers (D'Eon et al. 2002, DeCesare et al. 2003, D’Eon 2003, Yamazaki et al. 2008, Quaglietta et al. 2012). Subterranean depth, as we observed, can also cause GPS fix-rate bias given that fewer fixes are acquired at increased depths. Extra power is consumed when the GPS logger is unable to quickly acquire a satellite fix at the programmed logging interval. Therefore, individuals spending a greater time deep underground in their burrows are more likely to drain their batteries faster than individuals spending more of their time on or near the surface. This variability among individual gopher tortoises made determining exactly when we should swap out batteries difficult to predict accurately. With our initial programming schedule to attempt to collect a GPS location fix once every 30min, we obtained GPS data for ~3-4 months on average which required three GPS logger swaps per year in order to not lose data due to battery exhaustion. After adjusting the logging interval to attempt a fix hourly, battery life increased to ~5-6 months, requiring only two logger swaps per year. This protocol also decreases the amount of field effort required of the researcher and reduces handling time and stress on animal subjects in spatial and behavioral research.

While FSR is important to consider, the LE of GPS fixes is a more obvious concern. Some studies have tested GPS loggers while they were in motion on along routes as well as at stationary surface locations (Recio et al. 2011, Forin-Wiart et al. 2015). Additionally, canopy coverage has also been demonstrated to have a substantial effect on MLE (D’Eon et al. 2002, DeCesare et al. 2005, Lewis et al. 2007, Frair et al. 2010). However, to the best of our knowledge, no information was in the literature regarding subterranean GPS performance testing. Our stationary validation study provided us with realistic expectations in terms of MLE regarding the accuracy of these modified GPS loggers within gopher tortoise burrows at ecologically relevant depths. The range of MLE we observed at increasing depths (17.32-72.42m) is not surprisingly greater than the MLE observed on surface units in other studies such as Forin-Wiart et al. (2015) which reported a LE of 15.4m from similar low-cost GPS units. Another study which used professional wildlife-specific GPS loggers on Mojave Desert tortoises (*Gopherus agassizii*) reported a MLE of 8.6m (range of 3-38) in open desert habitat (Peaden et al. 2017). Canopy coverage at our site was assumed to be fairly consistent across the site due to the stationary locations all being within similar aged young longleaf pine silviculture stands; therefore, we attribute the observed differences in MLE to differences in depth.

While GPS loggers are advantageous in generating large amounts of data, the data are so numerous that dealing with large spatial datasets can be cumbersome without sufficient processing power. We also recommend setting *a priori* standards for which levels of data quality and quantity warrant inclusion in the targeted statistical analysis. In some cases, it may be appropriate to do more extensive data screening beyond filtering out GPS fixes erroneously recorded outside of deployment periods and extreme erroneous GPS outlier fixes (such as those recorded more than 5km from the study site as with our case). Several researchers have implemented various spatial weighting methods to reduce biologically “impossible” movement fixes due to GPS error and habitat features that may adversely affect the MLE or FSR (Frair et al. 2010, Webb et al. 2013, Lewis et al. 2017).

Home range analysis methods, such as nearest-neighbor k-LoCoH, can be effective methods to further filter data in the analysis stage to partially account for MLE in GPS data, such as is produced by increased subterranean depth. This compensation is accomplished by calculating inclusive nearest neighbor polygons for each individual location fix and then creating user-specified isopleths from all of those polygons. This process ensures that extreme and sparse outlier data for a given individual are automatically excluded and not as likely to have an effect of the resulting home range isopleths, a common sensitivity with some of the other spatial analyses methods. Due to the nature of large GPS logger data sets with a variable LE for each location fix, methods such as k-LoCoH that operate in this manner are appropriate (Getz and Wilmers 2004, Getz et al. 2007). However, it is still safest to be conservative in the analysis of spatial data to avoid false conclusions which might actually be due to error rather than biologically driven variability. For example, we saw that the 50% k-LoCoH method kept the isopleths error small enough during stationary testing that we felt comfortable using that same isopleth percentage to identify core home range areas in our tortoise data sets. In contrast, the 95% k-LoCoH method had a much larger error isopleth and would potentially overestimate the area being used, yet, that is still a relatively conservative approach to home range estimation. In contrast, the MCP method with GPS logger data was a gross overestimation of home range due to the inclusion erroneous outlier fixes. Therefore, with the help of advanced spatial analysis techniques such as the k-LoCoH, valuable information and sound conclusions can be drawn.

The ZoaTrack.org platform was particularly appealing due to its user friendliness being a Graphical User Interface program and the computational power provided via their cloud-based R and Google Earth interface. In most cases, the adehabitat R script ran the analyses smoothly through ZoaTrack provided the data were formatted correctly. We did encounter several glitches which returned errors that were determined to be inherent to the R code rather than ZoaTrack. An acceptable work-around in those few instances was to slightly increase the value of the k parameter; hence, we report a reasonable range of k-values (30-38) that we applied. While this type of adjustment is not typically applied with this estimator, we deemed it to be an option that was inconsequential to the isopleth construction; due to the amount of data clustered in the medium- and high-use areas of the home range, the final isopleth results were nearly identical and were biologically relevant based on our field knowledge and experience in tracking them via the VHF technology. While commercially available wildlife GPS loggers continue to improve, the units used in this study and other modified recreational GPS loggers may be able to provide affordable technology that generates biologically valuable data.

CHAPTER 5

CONCLUSION

This study found no statistically significant difference of home range size between resident and translocated gopher tortoises during ~5.5 months of monitoring post-release from temporary holding pens at the recipient site. We did however observe that throughout the cumulative study (including both the penning and post-penning periods), there was a statistically significant effect confirmed by three different spatial analysis techniques whereby translocated individuals used larger home ranges than that of resident tortoises during the same time period. This result could be useful in terms of the prioritization of recipient sites for future translocations based on the availability of suitable habitat not only at the point of release but also in the surrounding area at selected recipient sites for translocated tortoises to wander into without entirely leaving the area or being killed in that pursuit. Based on the lack of a statistically significant difference among residents and translocated individuals' home range size during the post-penning period, we feel that penning at the recipient site prior to release did help to establish site fidelity among translocated tortoises.

Interestingly, this study did not confirm the presence of statistically significant differences between sexes and home range size post-penning by using GPS logger spatial analysis techniques. However, the use of MCPs with VHF telemetry data indicated a statistically significant difference in sexes that was consistent with the frequently reported observation of male gopher tortoises using larger areas than that of females. Also, this study demonstrated that modified recreational GPS loggers have the capacity to perform under ecologically relevant conditions in order to effectively study gopher tortoise spatial ecology. Based on the various results emerging from this study, it appears there is still more to learn regarding the complex spatial ecology of gopher tortoises.

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APPENDICES

A. GOPHER TORTOISE CAPTURE DATASHEET.

Gopher Tortoise Capture Datasheet

ID: ____________
Accession #:_____________

Date (mm/dd/yy) _____ _____ _____ Collected by: ____

Time of capture (24-hr) ____:____ Recorded by:____

Capture Site	
☐ Priority Area 3: ________	☐ Priority Area 4
☐ Penholoway	☐ Other: ________________
UTM (NAD83): ________________, __________________	**Waypoint:** ______________

Recapture?	**Capture Method**	**Notes**
☐ Yes ☐ No	☐ Hand – on surface ☐ Bucket/Cage trap (burrow # _______) ☐ Excavated	

Excavation Tools Used (check all that apply)	**Burrow Notes**
☐ Hand pulled out ☐ Shovel ☐ Excavator ☐ Other: ___________________________	Burrow Size: ___________________________ Burrow Appearance: ______________________ Burrow Length:__________ Depth:__________ (surface to end chamber) Collapsed? ☐ Yes ☐ No

Release Location	
Date (mm/dd/yy)**:**___________________________ **Time** (24hrs.)**:**_____________________________	**Site name:** ____________________________ **Waypoint**: ______________ **Burrow #**: ______________ **UTM (NAD83):** ______________________, ______________________

1

Gopher Tortoise Capture Datasheet

ID: ______________
Accession #:______________

Date (mm/dd/yy) _____ _____ _____

Measured by: ____________________________ Recorded by: _______________________

Measurements

Sex/size class: ☐ Hatchling ☐ Juvenile ☐ Subadult ☐ Adult Male ☐ Adult Female

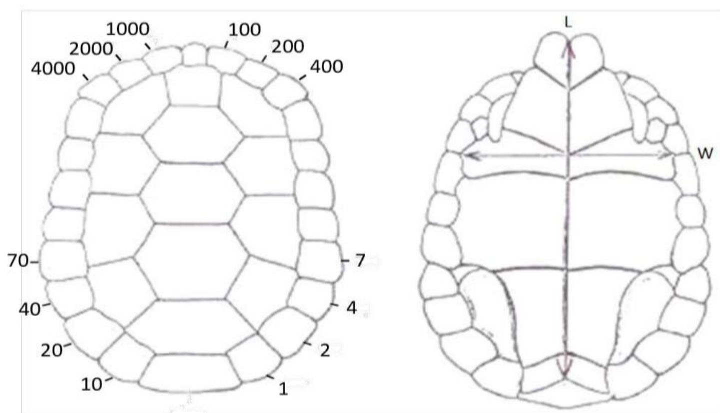
Criteria (CL):	Hatchling	Juvenile	Subadult	Adult Male	Adult Female
	< 68 mm Still in its 1st year	< 130 cm Scutes still yellow	≥ 130 and < 230 mm Plastron not concave	≥ 180 mm Plastron concave	≥ 230 mm Plastron not concave

CL (notch-notch) ________ mm Width (max) ________ mm Height (max) _______ mm

Gular Length ________ mm Weight (kg) at capture ________kg Weight (kg) w/ new equipment ______kg

Notch Code ________ No. Annuli ________

Marks



Circle which scutes are marked and draw position of marks on each scute (Check one):

☐ ID certain

Redrilled? yes no

☐ ID uncertain

Other possible IDs: ________________

Add-ons: (Check all that

Add-ons: (Check all that apply)

☐ Transmitter (Freq:_____________)

☐ Logger

☐ iButton (Serial:____________)

Took photos (Dorsal, ventral, & anterior)? yes no

Note any shell deformities or other distinguishing characteristics

B. CUMULATIVE SUMMARY OF GPS AND VHF SPATIAL DATA BY INDIVIDUAL

Individual GPS and VHF home range data for the cumulative study period (10 March 2016 to 23 November 2017). The GPS Fixes column shows only the successful fixes used in the analyses and does not include unsuccessful fix attempts. The VHF Tracks column shows the number of times an individual was manually radio-tracked during the study. Asterisks (*) denote the 28 individuals for which post penning period and penning period data were analyzed. Double asterisk (**) denote the two individuals for which no GPS logger data were acquired and therefore excluded from analyses.

Tortoise ID	Sex	Residency Status	Area (Ha) of 95% k-LoCoH	Area (Ha) of 50% k-LoCoH	Area (Ha) of VHF 100% MCP	# GPS Fixes	# VHF Tracks
*GT50	♀	Resident	0.71	0.01	1.27	3,174	57
GT54	♀	Resident	1.33	0.01	—	1,146	44
*Pandora44	♀	Resident	1.60	0.05	0.46	5,731	60
*GT52	♀	Resident	1.60	0.02	0.79	2,735	53
TheZika184	♀	Resident	1.65	0.02	—	849	41
*Lucky48	♀	Resident	1.73	0.05	1.85	5,682	57
*Athena182	♀	Resident	1.82	0.03	0.06	2,969	41
*GT189	♀	Resident	2.63	0.06	1.81	3,404	39
*GT183	♀	Resident	2.72	0.07	0.14	2,837	38
*GT192	♀	Resident	3.21	0.12	0.51	2,729	38
GT191	♀	Resident	3.83	0.03	1.62	745	42
GT181	♂	Resident	0.59	0.03	1.18	1,105	41
Phelps187	♂	Resident	0.62	0.02	0.21	493	41
*Adam49	♂	Resident	0.99	0.03	1.38	6,926	54
*GT190	♂	Resident	2.03	0.08	3.28	3,145	40
*GT55	♂	Resident	2.43	0.03	1.15	1,959	44
*Waldo47	♂	Resident	3.15	0.06	6.16	3,355	57
*Houdini53	♂	Resident	3.97	0.10	2.81	2,819	46
Yoshi56	♂	Resident	4.60	0.07	22.46	6,374	46
GT186	♂	Resident	4.80	0.13	0.70	838	38
Onyx135	♀	Translocated	2.04	0.03	4.76	1,248	37

*Peridot117	♀	Translocated	2.27	0.06	0.34	3,541	46
*Sapphire121	♀	Translocated	2.51	0.08	0.89	2,979	47
*Emerald106	♀	Translocated	3.11	0.04	1.72	4,377	45
Silica24	♀	Translocated	3.13	0.10	4.86	9,422	73
*Zircon37	♀	Translocated	3.18	0.04	9.40	4,410	46
*Basalt6	♀	Translocated	3.49	0.11	6.17	5,761	76
*Ruby124	♀	Translocated	3.87	0.06	2.81	4,563	46
Opal46	♀	Translocated	4.18	0.03	1.05	3,539	48
*Jade114	♀	Translocated	4.22	0.05	1.34	4,495	46
*Melanie145	♀	Translocated	4.73	0.11	7.90	4,790	36
*Pearl116	♀	Translocated	12.33	0.09	12.29	4,039	47
**Amythste105	♀	Translocated	—	—	11.80	—	39
**Topaz139	♀	Translocated	—	—	1.13	—	40
*Gypsum5	♂	Translocated	1.06	0.05	0.66	7,333	72
Quartz40	♂	Translocated	1.70	0.06	0.75	3,425	30
*Feldspar34	♂	Translocated	2.20	0.14	9.96	7,156	77
*Thorium35	♂	Translocated	3.78	0.05	10.71	5,549	73
*Titanium38	♂	Translocated	4.12	0.10	13.80	2,616	73
*Mica30	♂	Translocated	13.98	0.22	66.60	7,860	53