Comparison of methods for estimating abundance of gopher tortoises

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Abstract. Estimates of abundance of threatened and endangered species are crucial for monitoring population status and recovery progress. For most wildlife species, multiple abundance estimation methods are available and the choice of method should depend on cost and efficacy. We field-tested the cost and efficacy of line transect, total count, sample count, and double observer methods for estimating abundance of gopher tortoise (*Gopherus polyphemus*) burrows in two habitats that differed in vegetation density (sparse and dense) at the Ordway-Swisher Biological Station in north-central Florida. In the dense vegetation stratum, density of burrows estimated using the line transect method (8.58 ± 0.94 burrows ha^{-1}) was lower than that obtained from the total count method (11.33 burrows ha^{-1}). In the sparse vegetation stratum, estimated burrow density using the line transect method (11.32 ± 1.19 burrows ha^{-1}) was closer to the burrow density obtained from the total count method (13.00 burrows ha^{-1}). Density of burrows estimated using the double observer method was identical to that obtained from the total count method in dense vegetation stratum, but slightly greater than that obtained from the total count method in sparse vegetation stratum. Density of burrows estimated using the sample count method varied widely depending on the proportion of plots sampled. The cost of sampling as well as estimates of burrow density varied with habitat type. The line transect method was the least costly of the methods, and we were able to sample a larger effective area with the same effort. Using burrow cameras and patch occupancy modeling approach, we also estimated the probability of burrow occupancy by gopher tortoises (active: 0.50 ± 0.09; inactive: 0.04 ± 0.04), and used these values to estimate abundance of gopher tortoises. Using estimates of burrow abundance based on the line transect method, density of gopher tortoises was 2.75 ± 0.74 ha^{-1} in the sparse vegetation stratum. We recommend that gopher tortoise monitoring programs use rigorous methods for estimating burrow abundance (e.g., line transect methods) and the probability of burrow occupancy by gopher tortoises (e.g., patch occupancy modeling approach).

Key words: Abundance estimation; burrow density; burrow occupancy rate; double observer method; Florida; gopher tortoise; *Gopherus polyphemus*; line transect; patch occupancy model.

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Introduction

Abundance estimates are a prerequisite for listing or delisting of a species, and for monitoring recovery progress (Seber, 1982; Cassey and Mcardle, 1999; Williams et al., 2002). Furthermore, estimates of abundance are needed for understanding density-dependent relationships, for parameterizing and evaluating population models, and for formulating or evaluating management programs (Williams et al., 2002).

The gopher tortoise (*Gopherus polyphemus*) is a species of conservation concern in the southeastern US. It is federally listed as a threatened species in the western portion of its range (western Alabama, Mississippi, and Louisiana) (Lohoefener and Lohmeier, 1984; Federal-Register, 1987). In Florida, gopher tortoise populations have been declining for some time (Auffenberg and Franz, 1982; Schwartz and Karl, 2005; McCoy et al., 2006), and the species has recently been approved for reclassification to Threatened pending approval of a species management plan (FFWCC, 2006). Several state and federal agencies in the gopher tortoise range are charged with monitoring their status and population trends which require reliable estimates of abundance.

Estimating abundance of gopher tortoises is a two step process: estimation of burrow abundance and estimation of burrow occupancy rates. Gopher tortoises spend much of their time in the shelter of self-constructed underground burrows (Wilson et al., 1994), and direct observation of tortoises is difficult. These burrows are relatively easy to see due to their half-moon shape and large mound of sand (commonly referred to as the apron) at the burrow entrance. Because gopher tortoises are rarely seen outside their burrows, researchers typically estimate abundance of burrows, and frequently use it as an index of tortoise abundance (Cox et al., 1987; Smith et al., 2005; McCoy et al., 2006). The most commonly used methods for estimating the abundance of gopher tortoise burrows include line transect, total count, and sample count methods (Doonan, 1986; Mann, 1993; Epperson, 1997; Doonan and Epperson, 2001).

A second issue involved in the estimation of gopher tortoise abundance is that not every burrow is occupied by tortoises; there are typically more burrows than gopher tortoises and the number of burrows does not always correspond to the number of tortoises (McRae et al., 1981; Diemer, 1992; Smith et al., 1997; Eubanks et al., 2003; McCoy et al., 2006). Thus, an important issue relevant to gopher tortoise abundance estimation is the burrow occupancy rate (probability that a burrow is occupied by a gopher tortoise) (Diemer, 1992). Gopher tortoise abundance estimates are then obtained as the product of burrow occupancy rates and estimated burrow abundance. Auffenberg and Franz (1982) reported that 61.4% of all burrows (active and inactive) were occupied in their study. Some studies have used this or other similar values as a 'correction factor' to convert the burrow abundance into an estimate of tortoise abundance (e.g. Kushlan and Mazotti, 1984; Doonan, 1986; Doonan and Epperson, 2001; FFWCC, 2006; Gregory et al., 2006). Burrow occupancy rates vary over time and space, and unreliable estimates of occupancy rates can lead to erroneous estimates of gopher tortoise abundance

(Burke and Cox, 1988; Breininger et al., 1991; McCoy and Mushinsky, 1992; Moler and Berish, 2001).

Methods of estimating abundance of gopher tortoise burrows vary with respect to efficacy and cost, and rigorous field tests of these methods are needed to evaluate the efficacy relative to costs. Moreover, recent advances in occupancy models (MacKenzie et al., 2002; MacKenzie et al., 2006) offer the possibility of statistically rigorous estimates of burrow occupancy rates which were not possible previously.

Our objectives were to (1) investigate cost and efficacy of line transect, total count, sample count, and double observer methods for estimating abundance of gopher tortoise burrows, and (2) estimate the probability of burrow occupancy by gopher tortoises using burrow cameras and a patch occupancy modeling approach. We then combined estimates of burrow abundance with estimates of the probability of burrow occupancy to estimate abundance of gopher tortoises in the Ordway-Swisher Biological Station, Florida.

Materials and Methods

Study area

This study was conducted in the Ordway-Swisher Biological Station (http://www.ordway.ufl.edu) located in Putnam County, Florida (29°41′N and 82°W) (fig. 1) in the fall of 2005. The Biological Station encompasses approximately 4000 ha, and offers over 1600 ha of potential gopher tortoise habitat with old fields, pine plantations, and sand hill habitats of several burn frequency categories.

We selected a portion of the Ordway-Swisher Biological Station and stratified it into two strata (G5 and C3/C7) based on habitat maps, burn history and visual observation. Stratum G5, comprising of management unit G5, covered an area of approximately 110.3 ha, and was last burned in 2003 (two years before this study was conducted). Stratum C3/C7, comprising of management units C3 and C7, covered an area of approximately 116.5 ha, and was last burned in Feb 2005 (same year as this study). Due to the recent burn, stratum C3/C7 was more open with less dense vegetation than stratum G5 (R.R. Carthy, unpublished data). The study was conducted in two strata to investigate whether the probability of burrow detection, estimates of burrow abundance, and cost of the methods differed between the sandhill habitats with relatively high versus low vegetation density (Buckland et al., 2001).

Line transect method

Pilot study. We conducted a pilot study in the Ordway-Swisher Biological Station to estimate the transect length needed for robust estimates of burrow abundance using methods described in Buckland et al. (2001). We estimated that, to achieve a coefficient of variation of $\leqslant 15\%$ we needed to sample approximately 8 km of total transects in each stratum.

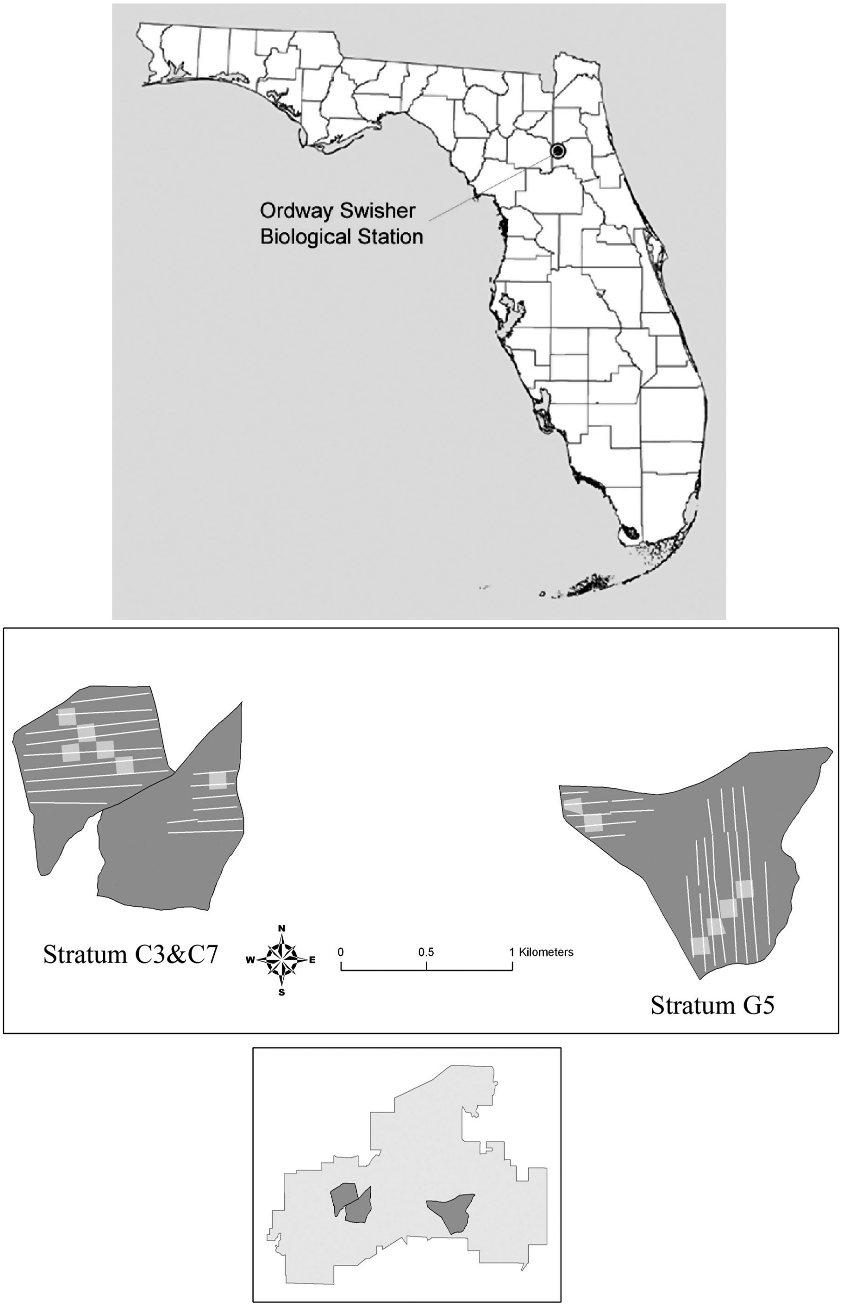

Figure 1. Map of Ordway-Swisher Biological Station in north-central Florida, USA, depicting stratum G5 and stratum C3/C7, and locations of line transects and plots.

Data collection. We placed 8 km of transects in each stratum systematically at predetermined distances (fig. 1). We allowed sufficient spacing (30-60 m) between transects to ensure that burrows would not be double-counted, while providing an adequate sample size for statistically robust results (Buckland et al., 2001). We oriented transects so that they did not run parallel to roads or other linear topographical features (Buckland et al., 2001; Williams et al., 2002) because they can affect the distribution of gopher tortoises. We placed flags and recorded GPS coordinates at the origin and end of all transects.

We used a total of two teams to collect data using the line transect method. Once the first team completed sampling transects we used a second team to collect data only in stratum G5 using the same protocol. These data were used to test for inter-observer variability in detection probability and estimates of burrow abundance. Each team, consisting of an observer and an assistant, walked along each transect line. The observer identified all burrows detected and the assistant then measured the perpendicular distance from the transect line to the burrow (Buckland et al., 2001; Williams et al., 2002). Perpendicular distance was measured from the transect line to either the burrow's mouth or the beginning of the burrow apron, whichever was closest to the transect line. The assistant also recorded GPS coordinates for each burrow, measured burrow width 50 cm inside the burrow, and classified each burrow according to width as juvenile (<14 cm wide), sub-adult (14-23 cm wide), and adult (>23 cm) (Alford, 1980; Smith, 1992). The observer classified each burrow into one of two burrow status categories: active and inactive. Active burrows had burrow aprons and entrances with little or no debris, and had evidence of tortoise occupation. Inactive burrows, on the other hand, had debris and leaf litter on the apron, at the mouth, and in the burrow tunnel, and were unlikely to be occupied by a gopher tortoise. In some cases burrow mouths were degraded so that they did not have the classic, half-moon tortoise shape.

The assistant did not participate in detecting burrows but simply collected and recorded data for burrows detected by the lead observer. All detected burrows were temporarily marked with a numbered tag so as to avoid double counting of burrows. Tags were hidden from view of the second observer, but were placed in a consistent location at burrows so they were easily located by the second observer's assistant upon close examination of the burrows detected by the second observer.

One critical assumption of line transect sampling is that all objects located on the line transect are seen and recorded (Buckland et al., 2001; Williams et al., 2002). Burrows of adult and juvenile gopher tortoises are conspicuous, and are associated with mounds that are hard to miss from a close distance (Lohoefener, 1990; Doonan and Epperson, 2001). We are, therefore, confident that all tortoise burrows that were directly on the line were detected.

Data analysis. We used Program DISTANCE (Thomas et al., 2003) to analyze the line transect data to estimate abundance of gopher tortoise burrows. The program provides a flexible framework for parameterization and comparison of alternative models (Buckland et al., 2001). Prior to modeling, we excluded 5% of observations (furthest from the line) from the analysis to remove possible outliers (Buckland et al., 2001). We ran several parametric models, each consisting of a key function and a series adjustment term (Buckland et al., 2001). We used Akaike's Information Criterion (AIC) values for model comparison and identified the model with the Lowest AIC as most parsimonious; we reported estimates of abundance based on the most parsimonious model (Burnham and Anderson, 2002).

Of the parametric models discussed above, we selected the most parsimonious model that allowed covariates and tested for the effect of the width of the burrow entrance (in cm) on the detection probability for Observer 1 in both strata. We then compared the AIC values for models with and without burrow width as a covariate to evaluate the effect of burrow width on estimates of detection probability and burrow density. Using the same procedure, we used observer as a covariate to test for inter-observer variability in detection probability in stratum G5. Data for the two observers were pooled together to test for inter-observer variability, and the resulting AIC value was compared to the sum of the AIC values obtained from the separate analyses of data collected by the two observers without observer as a covariate (Buckland et al., 2001: 111).

Total count, sample count and double observer methods

Data collection. We conducted a total count of burrows in a portion of the area where transect data had been collected. We selected six 1-ha plots in each stratum. The four corners of the plots were flagged and their coordinates were determined using a GPS unit. We further subdivided the plots into ten 20 × 50 m subplots and each subplot was comprehensively searched for burrows by two observers so detectability could be estimated (Nichols et al., 2000; Williams et al., 2000). The observers recorded all pertinent information for detected burrows. Our effective sampling area in each stratum comprised of these six 1-ha plots. Sample count was the total count in a random sample of subplots.

We implemented the dependent double observer method following Nichols et al. (2000) and Williams et al. (2002). Each of the 20 × 50 m subplots was comprehensively searched for burrows by two dependent observers (primary observer and secondary observer, where the secondary observer is aware of the burrows detected by the primary observer). The primary observer surveyed the plots, and flagged and called out burrows detected to the secondary observer. The secondary observer recorded the information and proceeded to survey the plots to detect additional burrows (Nichols et al., 2000). At the completion of sampling of each subplot, data were comprised of burrows detected by the primary observer, and burrows missed by the primary observer but detected by the secondary observer. Observers alternated roles on consecutive subplots, as recommended by Nichols et al. (2000).

We conducted the total count, sample count and double observer methods on the same six 1-ha plots in each stratum. The field methods for the three methods were identical and data collected were the same. We used data collected for the total count method for the sample count calculations by selecting a subset of plots for which total counts were conducted (see next section).

Data analysis. The estimated abundance of burrows using total count was the total number of burrows detected by both observers. Estimates of cost and abundance obtained from the sample count method can vary depending on the proportion of total area sampled, spatial distribution of burrows and the choice of the sample plots. Thus, we evaluated the effects of selecting different proportions of plots on the estimate of burrow abundance by utilizing data collected for the total count method. We selected all possible combinations of 3, 4 and 5 out of the 6 plots; each of these plots was thoroughly surveyed by two observers as described previously. The number of burrows in the sampled plots was then extrapolated to obtain an estimate of the number of burrows to the entire 6-ha area sampled. Sampling 100% of the plots (6 out of 6) is the total count.

We used Program DOBSERV (Hines, 2000) to analyze the double observer data. The overall detectability was estimated as (Nichols et al., 2000):

$$\hat{p} = 1 - (x_{12}x_{21}/x_{22}x_{11}),$$

where $\hat{p}$ = estimate of overall detectability of both observers, x_{11} = number of burrows detected by observer 1 in a primary role, x_{21} = number of burrows detected by observer 2 in a primary role, x_{12} = number of additional burrows detected by observer 1 in a secondary role and x_{22} = number of additional burrows detected by observer 2 in a secondary role. This estimate of overall detectability ($\hat{p}$) was used to obtain the estimate for the population size for the sampling area ($\hat{N}$) by dividing the total number of burrows detected by all observers ($x_{..}$) by $\hat{p}$ (Nichols et al., 2000). The standard error ($\mathrm{SE}(\hat{N})$) and the 95% confidence interval for $\hat{N}$ were estimated using Program DOBSERV (Hines, 2000).

We divided $\hat{N}$ obtained from each method by the area of the study site sampled to estimate the burrow density ($\hat{D}$) (burrows ha^{-1}). We multiplied $\hat{D}$ by the area of the stratum to obtain an estimate of burrow abundance in each stratum.

Burrow occupancy rates

Data collection. To estimate the probability that a burrow was occupied by a gopher tortoise (burrow occupancy rate) we conducted burrow occupancy surveys in management unit C3 of stratum C3/C7 (fig. 1). We examined a sub-sample of 56 burrows from C3 that were marked during the total counts with a burrow camera on three consecutive days (beginning either in the morning or early afternoon) to determine occupancy status. This sub-sample contained both active and inactive burrows. We used the Econo GeoVision, Jr. camera system designed by Marks Products, Inc. (Williamsville, Virginia) for use in borehole and water well systems. We sanitized the equipment with diluted Nolvasan® after examining each burrow, to minimize the risk of disease transmission. We classified the burrows as 'empty' if the operator was certain that she/he had reached the end of the burrow and no gopher tortoise was present. Burrows were considered 'occupied' only if the operator could identify a gopher tortoise with absolute certainty. Burrow occupancy was considered 'undetermined' if the operator could not maneuver the camera to the end of the burrow due to burrow architecture (e.g., dramatic turns or tunnel size) or debris (e.g., leaf litter and/or sand). Burrows with undetermined occupancy status were not used in analysis of occupancy rates.

Data analysis. We used the occupancy data collected using a burrow camera and a statistically robust occupancy modeling approach (MacKenzie et al., 2002) implemented in Program MARK (White and Burnham, 1999) to estimate detection probability and burrow occupancy rate. We assumed that occupancy status remained unchanged between sampling occasions. Occupancy survey was conducted as described previously. A burrow was considered occupied by a tortoise (coded 1) if the observer was certain that a tortoise was present; it was considered unoccupied (coded 0) if the observer was certain the burrow was not occupied. Using these occupancy data collected over 3 sampling occasions, we fitted the patch occupancy model (MacKenzie et al., 2002; MacKenzie et al., 2006). We used AIC to select the most parsimonious model. Using the most parsimonious model identified, we tested for the effect of the width of the burrow entrance (in cm) on the detection probability and the occupancy rate by modeling the logit of each rate as a linear function of burrow width. If the 95% confidence interval for the slope parameter (β) did not include 0, the relationship was considered statistically significant (Williams et al., 2002).

Costs

We recorded the time taken for data collection for each method in person-hours. The time spent collecting data is commonly the greatest cost incurred during a study, and therefore a good indicator of the cost effectiveness of a method. The amount of time needed for analysis of data varies among individuals depending on mathematical background, computer skills, and learning curves, and thus are not reported. The costs of equipment required for analysis may also vary tremendously and thus are not reported.

Results

Line transect

A total of 28 line transects was placed in stratum G5 with a total length of 8025 m. Observer 1 detected 163 burrows and Observer 2 detected 150 burrows. For Observer 1, the model with the lowest AIC was Uniform Cosine (table 1). Based

Table 1. Comparison of models fitted to line transect data. For each model the difference in Akaike's Information Criterion values (ΔAIC); and the number of parameters are presented.

		Observer 1		Observer 2[b]	
Stratum	Model	ΔAIC	Parameters	ΔAIC	Parameters
G5					
	Uniform cosine[a]	0.00	1	0.00	1
	Half normal cosine	0.13	1	0.93	1
	Half normal hermite	0.13	1	0.93	1
	Uniform simple polynomial	1.46	2	0.98	2
	Hazard rate cosine	2.22	2	2.31	2
	Hazard rate simple polynomial	2.22	2	2.31	2
C3/C7					
	Hazard rate cosine[a]	0.00	2	–	–
	Hazard rate simple polynomial	0.00	2	–	–
	Uniform cosine	0.98	2	–	–
	Half normal cosine	1.32	2	–	–
	Half normal hermite	3.14	1	–	–
	Uniform simple polynomial	3.59	3	–	–

[a] Most parsimonious model.
[b] Data collected by Observer 2 in stratum C3/C7 were not analyzed.

on this model, the estimated burrow density (±SE) was 8.58 ± 0.94 burrows ha^{-1} ($cv = 11.0\%$). For Observer 2, the model with the lowest AIC was also Uniform Cosine (table 1). Based on this model, the estimated burrow density was 8.49 ± 0.98 burrows ha^{-1} ($cv = 11.5\%$) (table 2).

A total of 16 line transects was placed in stratum C3/C7 with a total length of 8003 m. The first observer (Observer 1) detected 262 burrows. For Observer 1, the models with the lowest AIC were Hazard Rate Cosine and Hazard Rate Simple Polynomial (table 1). The results for these two models were identical, so we selected the Hazard Rate Cosine model. Based on this model, the estimated burrow density was 11.32 ± 1.19 burrows ha^{-1} ($cv = 10.5\%$) (table 2).

In stratum G5, we evaluated the effect of burrow width on detection probability for Observer 1 using the Half Normal Cosine model. The difference between the probability of detecting smaller burrows and the probability of detecting larger burrows was not substantial (AIC for model with burrow width as a covariate: 475.86; AIC for model without burrow width as a covariate: 477.87). Consequently, estimates of burrow density were very similar (with burrow width as a covariate: 8.99 ± 1.06 burrows ha^{-1}; without burrow width as a covariate: 8.58 ± 0.94 burrows ha^{-1}). In stratum C3/C7, we evaluated the effect of burrow width on detection probability for Observer 1 using the Hazard Rate Cosine model. Although burrow width seemed to influence detection probability (AIC for model with burrow width as covariate: 867.97; AIC for model without burrow width as covariate: 957.48), the difference in the estimate of burrow density was not substantial (with

Table 2. Overall summary of estimates of abundance of gopher tortoise burrows for each abundance estimation method in two strata (G5 and C3/C7), Ordway-Swisher Biological Station, Florida. $\hat{D}$ is the estimated burrow density (with 95% confidence interval) in burrows ha^{-1} (range of estimated burrow density is provided for the sample count method), and $\hat{N}(tot)$ is the estimated number of burrows in the stratum. Cost is presented in terms of time (person-hours) needed to sample 1 ha.

	Method		$\hat{D}$	$\hat{N}(tot)$	Cost
G5					
	Line transect[a]				
		Obs 1	8.58 (6.87-10.73)	946.40 (757.60-1183.72)	0.52
		Obs 2	8.49 (6.73-10.71)	936.25 (742.16-1181.06)	–
	Sample count				
		50%[b,c]	8.00-14.66	882.21-1616.66	1.19
		66%[b,c]	9.00-13.50	992.49-1488.74	1.57
		83%[b,c]	10.00-12.40	1102.77-1367.43	1.98
	Total count	100%[b]	11.33	1249.44	2.38
	Double observer		11.33	1249.44	2.38
C3/C7					
	Line transect[a,d]				
		Obs 1	11.32 (9.19-13.94)	1318.31 (1070.35-1623.57)	0.46
	Sample count				
		50%[b,c]	10.67-15.33	1246.21-1781.97	1.04
		66%[b,c]	11.50-14.50	1339.39-1688.79	1.38
		83%[b,c]	12.20-13.80	1420.92-1607.27	1.73
	Total count	(100%[b])	13.00	1514.09	2.08
	Double observer		13.04 (13.00-13.55)	1518.75 (1514.48-1578.73)	2.08

[a] Results are based on the most parsimonious model (table 1).
[b] Proportion of plots sampled.
[c] Range of estimates of density.
[d] We did not analyze line transect data for Observer 2 for stratum C3/C7 because Observer 2 did not collect data independently of Observer 1.

burrow width as covariate: 11.26 ± 0.84 burrows ha^{-1}; without burrow width as covariate: 11.32 ± 1.19 burrows ha^{-1}).

Additionally, in stratum G5, we evaluated inter-observer variability in detection probability using pooled data collected by the two independent observers and the Half Normal Cosine model. There was no evidence for difference in detection probability between the two observers (AIC for model with pooled observations: 1871.19; sum of AIC values for models analyzed separately for Observer 1 and Observer 2: 1871.21), and the difference in the estimate of burrow density was not substantial (model with pooled observations: 8.55 ± 0.71 burrows ha^{-1}; model analyzed separately for Observer 1: 8.68 ± 0.97 burrows ha^{-1}; model analyzed separately for Observer 2: 8.41 ± 1.01 burrows ha^{-1}).

Total count, sample count and double observer

The total number of burrows in the six 1-ha plots sampled was 68 (11.33 burrows ha^{-1}) in stratum G5 and 78 (13.00 burrows ha^{-1}) in stratum C3/C7 (table 2). The abundance of burrows based on the sample count method varied widely in both strata based on the proportion of sample plots used (table 2). In stratum G5, when 50%, 66% and 83% of the plots were sampled, the density of burrows in the sampling area ranged from 8.00 to 14.66, 9.00 to 13.50 and 10.00 to 12.40 burrows ha^{-1}, respectively. In stratum C3/C7, when 50%, 66% and 83% of the plots were sampled, the density of burrows in the sampling area ranged from 10.67 to 15.33, 11.50 to 14.50, and 12.20 to 13.80 burrows ha^{-1}, respectively.

The overall detectability ($\hat{p}$) estimated using the double observer method was 1.0, and $x_{..}$ and $\hat{N}$ were 68 (11.33 burrows ha^{-1}) in stratum C3/C7; $\hat{p}$ was 0.997 ± 0.003, $x_{..}$ was 78 and $\hat{N}$ was 78.23 ± 0.53 (13.04 burrows ha^{-1}) in stratum G5 (table 2). Because $\hat{p}$ was 1.000, SE($\hat{N}$) or 95% confidence interval could not be estimated for stratum C3/C7 (table 2).

Burrow occupancy rates and abundance of gopher tortoises

The most parsimonious model (table 3) indicated that detection probability (probability of observing a gopher tortoise if it was in the burrow) was 0.92 ± 0.04 and did not differ between burrows classified as active or inactive. However, the occupancy rates were significantly different between the two groups (active: 0.50 ± 0.09; inactive: 0.04 ± 0.04). There was no evidence that width of the burrow entrance influenced the occupancy rate or the detection probability.

Using the occupancy rates for active and inactive burrows, and based on the proportion of active and inactive burrows we estimated the density of gopher tortoises in stratum C3/C7. The estimated tortoise density varied from 2.19 ± 0.43 to 3.14 ± 0.61 tortoises ha^{-1} (active and inactive burrows) depending on the method used, and in the case of sample count, depending on the proportion of plots sampled (table 4).

Table 3. Comparison of models fitted to patch occupancy data to estimate burrow occupancy rate. For each model the difference in Akaike's Information Criterion values (ΔAIC), the number of parameters, and the model weight are presented. 'p' is the detection probability, 'psi' is the occupancy rate, 'g' indicates the group effect (active/inactive burrows), and a period (·) indicates a constant parameter model.

Model	ΔAIC	Parameters	Model weight
{p(·) psi(g)}[a]	0.00	3	0.71
{p(g) psi(g)}	1.81	4	0.29
{p(g) psi(·)}	12.88	3	0.01

[a] Most parsimonious model.

Table 4. Estimated density (±SE; ha^{-1}) of gopher tortoises in stratum C3/C7, Ordway-Swisher Biological Station, Florida. $\hat{D}_B$ (Active) and $\hat{D}_B$ (Inactive) are estimates of density of active and inactive burrows in the stratum, $\hat{D}_{GT}$ (Active) and $\hat{D}_{GT}$ (Inactive) are estimates of density of gopher tortoises in active and inactive burrows, respectively, and $\hat{D}_{GT}(tot)$ is the estimated density of gopher tortoises in all burrows in the stratum.

		Burrow density		Gopher tortoise density[a]		
Method		$\hat{D}_B$ (Active)	$\hat{D}_B$ (Inactive)	$\hat{D}_{GT}$ (Active)	$\hat{D}_{GT}$ (Inactive)	$\hat{D}_{GT}(tot)$
Line transect[b]	Obs 1	5.00 ± 0.53	6.32 ± 0.66	2.50 ± 0.47	0.25 ± 0.28	2.75 ± 0.74
Sample count	50%[c]	3.84-5.49	6.86-9.81	1.92 ± 0.15-2.75 ± 0.22	0.27 ± 0.27-0.39 ± 0.39	2.19 ± 0.43-3.14 ± 0.61
	66%[c]	4.13-5.21	7.37-9.29	2.07 ± 0.17-2.61 ± 0.21	0.29 ± 0.29-0.37 ± 0.37	2.36 ± 0.46-2.98 ± 0.58
	83%[c]	4.38-4.95	7.82-8.85	2.19 ± 0.18-2.48 ± 0.20	0.31 ± 0.31-0.35 ± 0.35	2.50 ± 0.49-2.83 ± 0.55
Total count	100%	4.67	8.33	2.34 ± 0.19	0.33 ± 0.33	2.67 ± 0.52
Double observer		4.68 ± 0.03	8.36 ± 0.06	2.34 ± 0.20	0.33 ± 0.34	2.67 ± 0.54

[a] Standard error for gopher tortoise density was calculated using the delta method (Williams et al., 2002).
[b] The proportion of active and inactive burrows detected using the line transect method for Observer 1 was 0.44 and 0.56, respectively. The proportion of active and inactive burrows detected using the total count methods was 0.36 and 0.64, respectively.
[c] Range of estimates of density.

Costs

The cost of sampling varied from 0.52-2.38 person-hours ha^{-1} in stratum G5, and from 0.46-2.08 person-hours ha^{-1} in stratum C3/C7, depending on the methods used, and in the case of sample count, depending on the proportion of plots sampled (table 2). We did not analyze cost of line transect data collected by Observer 2, because Observer 2 did not place transects or measure and record information for burrows that had already been detected by Observer 1. Costs of implementing the double observer method were identical to the cost for the total count method.

The cost of sampling burrows with a burrow camera to determine occupancy status was 0.16 person-hr per burrow camera observation. A total of 168 burrow camera observations were performed (three observations for each of the 56 burrows scoped) requiring a total time of 26.88 person-hrs.

Discussion

Comparison of abundance estimation methods

To monitor population status, and for appropriate recovery efforts for gopher tortoises, reliable estimates of abundance are needed. Methods that are currently used to estimate abundance of gopher tortoises vary with respect to statistical rigor, efficacy and cost. Given the pivotal role of gopher tortoises in ecosystems where they are found (Eisenberg, 1983; Wahlquist, 1991), it is essential to use rigorous yet cost-effective methods for estimating and monitoring tortoise abundance.

We field-tested the efficacy and cost of line transect, total count, sample count and double observer methods for estimating abundance of gopher tortoise burrows. In the dense vegetation stratum (G5), the estimated burrow density using the line transect method for both observers (8.58 and 8.49 burrows ha^{-1}, respectively) was nearly 3 burrows ha^{-1} less than burrow density of 11.33 burrows ha^{-1} obtained from total count method. Estimates based on total count method did not fall within the 95% confidence intervals of those obtained from line transect method (table 2). In the sparse vegetation stratum (C3/C7), the burrow density estimated using the line transect method (11.32 burrows ha^{-1}) was closer to the burrow density obtained from the total count method (13.00 burrows ha^{-1}). The total count fell within the 95% confidence interval for estimates obtained from the line transect method (table 2).

Mann (1993) compared estimates of tortoise burrow abundance obtained from line transect and total count methods, and found that line transect method overestimated burrow abundance by as much as 49% in 13 sites and 32% on seven sites. Results from similar studies suggest a tendency for line transects to overestimate abundance when compared to total counts (Doonan, 1986; Epperson, 1997; Doonan and Epperson, 2001). Our results do not agree with findings that the line transect method tends to overestimate burrow abundance. In fact, estimates of burrow abundance obtained from the line transect method were lower than those obtained from total count in stratum G5; they did not differ significantly in stratum C3/C7 (table 2). These results suggest that the estimated burrow abundance obtained from the line transect method are not consistently greater or smaller than those obtained from the total count method. Therefore, the line transect method likely captured a greater amount of spatial variability in distribution and abundance of burrows in the study area. We also had a sufficiently large sample size for a reasonable coefficient of variation. Additionally, for the total count and double observer methods, we used $\geqslant 2$ observers to thoroughly search the sampling area, and ensured that detection probability could be estimated.

Consistent with previous studies (McCoy and Mushinsky, 1995; Epperson, 1997; Marques et al., 2001; Hermann et al., 2002; McCoy and Mushinsky, 2005), our estimates of burrow density varied with habitat type and burn frequency. Density estimates obtained from all methods were higher in stratum C3/C7 which had comparatively sparse vegetation and a higher burn frequency. The higher density of burrows and tortoises in stratum C3/C7 indicates that this stratum offered a better habitat for the tortoises as the more open canopy cover likely resulted in increased herbaceous ground forage and minimum light levels required by gopher tortoises (Auffenberg and Franz, 1982; Aresco and Guyer, 1999).

Burrow occupancy

Our estimates of burrow occupancy rates (active: 0.50 ± 0.09; inactive: 0.04 ± 0.04) were substantially lower than Auffenberg and Franz's 'correction factor' of 61.4% for active and inactive burrows (Auffenberg and Franz, 1982). Some studies have used this or a similar correction factor for converting estimates of burrow abundance to tortoise abundance (Kushlan and Mazotti, 1984; Doonan, 1986; Doonan and Epperson, 2001; FFWCC, 2006; Gregory et al., 2006). However, this approach ignores the spatial, temporal or habitat-specific variation in occupancy rate and can cause estimates of gopher tortoise abundance to be unreliable (Burke and Cox, 1988; Breininger et al., 1991; McCoy and Mushinsky, 1992; Moler and Berish, 2001). Moreover, our study is the first to apply the patch occupancy modeling approach (MacKenzie et al., 2002; MacKenzie et al., 2006) to estimate and model burrow occupancy rates. When appropriate data are available, this approach also provides framework for testing relevant biological hypotheses.

Because of time and resource limitations, we conducted burrow occupancy surveys only in management unit C3 of stratum C3/C7, and we did not have empirical estimates of burrow occupancy rates for stratum G5. Occupancy rates may vary among habitats due to the ecological needs of gopher tortoises, and habitat-specific estimates of occupancy rates should be used whenever possible. Estimates of occupancy rates may also be influenced by the season, time of the day when data are collected, and time interval between successive samples; these factors should be considered whenever possible. Additionally, there is the possibility of the same tortoise occupying more than one burrow during the burrow occupancy surveys, resulting in an overestimation of the occupancy rate. These potential problems can be minimized by appropriate sampling design. Nonetheless, we found that patch occupancy models (MacKenzie et al., 2002; MacKenzie et al., 2006) offer statistically robust approach to estimating burrow occupancy rates and should be considered in future studies.

Costs of implementation

The total count and sample count methods were relatively straightforward to implement, and required no sophisticated software for data analyses. However, these methods are costly, particularly when a substantial proportion of the sites need to be sampled. Moreover, the total count and sample count methods do not offer rigorous estimates of precision or meaningful approaches to statistical inference. The double observer method partially addressed some of these concerns by providing estimates of precision (when detectability is less than 1.0), but is costly to implement. Using the sample count method, the range of extrapolated estimates for burrow density became narrower as the sampling proportion increased (table 2). However, cost of sampling increased as more time was needed to collect the data (table 2).

The line transect method was the least costly of the methods, and we were able to sample a larger effective area with the same effort. The method is considered statistically rigorous and robust, provides statistical measures of precision, and offers a framework for statistical inferences (Buckland et al., 2001; Krzysik, 2002; Williams et al., 2002). However, the low cost of sampling in the field may be somewhat offset by increased costs of data analysis, as a good understanding of underlying theory, sampling protocol and working knowledge of Program DISTANCE is needed for effective implementation of this method.

Costs of data collection differed between the two strata in our study site. The sparse vegetation stratum (C3/C7) had a lower relative cost of sampling for all the abundance estimation methods than dense vegetation stratum. In our study, sample counts and total counts were substantially more costly than line transects in both strata. Detection time may be substantially less in sparse vegetation (Lohoefener and Lohmeier, 1984; Burke and Cox, 1988; Lohoefener, 1990; Diemer, 1992), and prescribed burns prior to sampling may further reduce cost of sampling (Smith, 1992; Mann, 1993; Moler and Berish, 2001).

Among other factors, the selection of an abundance estimation method should consider the habitat type of the study area, and available time and resources (Ellis and Bernard, 2005). With a stratified sampling design, and an adequate sample size, the line transect method is perhaps the most efficient method for estimating gopher tortoise burrow abundance because: (1) it is less costly than total and sample count methods, (2) it is more likely to capture a wider range of spatial variation in the distribution and abundance of burrows, (3) it offers statistically robust estimates of measures of precision and (4) provides a flexible framework for evaluating effects of covariates on estimates of abundance.

If one wishes to implement the total (or sample) count method, we recommend using multiple observers, and providing evidence that detectability was $\sim$1.0. We note, however, that the total count method does not provide an estimate of variance, nor does it provide a framework for statistical test of hypotheses. The double observer approach is reasonable if one wishes to implement a count-based method, but is unsure that detectability is equal to 1.0.

We recommend that burrow cameras (or similar technologies) should be employed, along with a patch occupancy modeling approach for data analysis, to estimate burrow occupancy rates and to test hypothesis regarding the occupancy rate or detection probability. If a study is conducted in >1 habitat types, we recommend obtaining habitat-specific estimates of occupancy rates. Finally, we suggest that gopher tortoise monitoring programs should simultaneously consider burrow abundance and burrow occupancy rates. This is because changes in gopher tortoise abundance may be reflected in changes in one or both of these parameters (i.e., burrow abundance and burrow occupancy rate), and changes in one may not be interpreted as an indicator of changes in tortoise abundance.

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