Comparison of Methods for Estimating Key Largo Woodrat Abundance

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Abstract: Monitoring abundance of the endangered Key Largo woodrat (*Neotoma floridana smalli*) is necessary to understand population responses to prescribed management actions. We compared efficiency of adaptive cluster sampling (ACS) and stratified-random sampling (SRS) for estimating Key Largo woodrat abundance and compared three stratification designs using poststratification. We established 40 trapping grids using a stratified random design and adaptively sampled around grids on which at least 1 individual was captured. We captured 11 individuals on 40 random grids and an additional 22 individuals on 33 adaptive grids. Despite the increased capture rate, ACS was less efficient than SRS with an estimator variance twice as large with equal sample sizes. Although poststratification effectively lowered estimator variance, our data suggest that attaining the required sample sizes to reliably estimate abundance likely will be cost-prohibitive. Monitoring of improved habitat at the patch scale along with representative controls may be more cost-effective for evaluating the success of prescribed management.

Key words: Abundance, adaptive cluster sampling, Key Largo woodrat, Neotoma floridana smalli, poststratification, stratified random sampling

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The Key Largo woodrat (Neotoma floridana smalli) historically ranged throughout the hardwood hammocks of Key Largo, Florida, but is now restricted to state and federally protected lands on the northern one-third of the island (Barbour and Humphrey 1982). Despite protection of its remaining habitat from development, the population has continued to decline after listing in 1984 (McCleery et al. 2006b). Recent research determined that the species currently occupies approximately 20% of the hardwood hammock on north Key Largo (McCleery et al. 2006b), suggesting that habitat quality has been severely degraded. The U.S. Fish and Wildlife Service listed habitat improvement as a primary management goal in the species' recovery plan (U.S. Fish and Wildlife Service 1999). Accordingly, identifying and evaluating appropriate sampling designs for monitoring Key Largo woodrat abundance are necessary to evaluate effectiveness of future habitat improvement efforts.

Accuracy and precision of population abundance estimates can be increased if trapping grids are initially allocated within biologically meaningful strata via stratified random sampling (SRS; Thompson 2002). Past Key Largo woodrat population estimates have been obtained using capture-recapture data from trapping grids that were not stratified (Barbour and Humphrey 1982, Humphrey 1988) or were stratified by hammock age (McCleery et al. 2006b). Recent research suggests that Key Largo woodrats are clustered around artificial denning substrate, such as rock or debris piles (McCleery et al. 2006b, Winchester et al. 2009). Given high frequency of use around artificial substrate, delineating areas in proximity to these resources might be an effective means of stratifying sampling effort. However, clustering of Key Largo woodrats around artificial substrate likely has resulted in an unevenly distributed population within available habitat.

Using traditional sampling designs (e.g., SRS) to sample unevenly distributed populations may result in imprecise estimates of abundance even with relatively extensive trapping effort (Williams et al. 2002). Adaptive Cluster Sampling (ACS) was developed as an alternative to traditional sampling designs to more effectively estimate population size of rare and clustered populations (Thompson 1990). In an ACS approach, additional sampling units are selected from all immediately surrounding units when a pre-defined criterion is met (e.g., when 1 individual is detected) resulting in a "neighborhood" of sampling units around the original unit. The process continues until the neighborhood is bounded by "edge units" that do not meet the criterion. ACS allows for unbiased estimates of population parameters when using information obtained in previously collected samples to update sampling probabilities in adaptively sampled units (Williams et al. 2002). ACS also can be used when the initial sample is selected using stratified random sampling (Thompson 1991).

Our objective was to compare efficiency of ACS to SRS for providing estimates of Key Largo woodrat abundance from capturerecapture data. We used estimates of detection rate and proportion of transient individuals to correct for bias in ACS and SRS estimates.

Study Area

Our study area consisted of upland forest habitat (hardwood hammocks) on Crocodile Lake National Wildlife Refuge and Dagny Johnson Key Largo State Botanical Preserve. Hardwood hammocks of this region are closed-canopy forest containing a diverse assemblage of evergreen and semi-deciduous tree and shrub species (U.S. Fish and Wildlife Service 1999). Canopy closure creates a shaded environment on the forest floor resulting in a sparse shrub and herb layer. As a result, the understory consists mainly of seedlings and saplings of canopy and sub-canopy species. Hammock habitat is bordered by mangroves along the coast with a relatively narrow transitional zone (Ross et al. 1992).

Methods

Sampling Design

We divided the 850-ha area of hardwood hammock into 75-x 75-m (0.56-ha) sampling grids (units) and selected 40 via a stratified random design. Because rock or debris piles are important substrates for Key Largo woodrat nest sites (McCleery et al. 2006b, Winchester et al. 2009), we defined our two strata as the center of the sampling unit being ≤ 75 or >75 m from a rock or debris pile. We obtained locations of all known rock and debris piles from park and refuge personnel and conducted searches as time allowed prior to sampling to identify additional locations. We recorded coordinates of all locations with a Global Positioning System (GPS; Trimble GeoXT) prior to selection of sampling units and created a layer in a Geographic Information System (ArcGIS; Environmental Systems Research Institute, Redlands, California) to delineate stratum boundaries. We selected sampling units in proportion to area available in each stratum, such that each unit had an equal inclusion probability.

Using the initial SRS, we set the criterion for adaptive sampling at one individual captured on a unit. When an individual was captured we sampled all immediately surrounding units with enough area in hardwood hammock habitat to contain a 50-x 50-m trapping grid (i.e, the neighborhood). We applied a stopping rule of one adaptive addition to each initial unit meeting the criteria to limit the final sample size. Sampling units added adaptively were allowed to cross stratum boundaries.

Trapping and Handling

We established a 3×3 trapping grid with 25 m between stations within each sampling unit. At each station we placed two 10.2-x 11.4-x 38.1-cm, vented Sherman traps (model PXLF15, H. B. Sherman Traps Inc., Tallahassee, Florida) with raccoon (*Procyon lotor*)proof door latches baited with peanut butter and crimped oats. We opened traps for four consecutive nights between 27 April and 1 June 2005, checking each trap daily within the first three hours after sunrise. All captured individuals were weighed, sexed, and marked with passive integrated transponders (PIT) tags (BioMark, Boise, Idaho) and No. 1005 Monel ear tags (National Band and Tag, Newport, Kentucky). We conducted all capture and handling under U.S. Fish and Wildlife Service Endangered Species Permit No. TE0959080-1, State of Florida Fish and Wildlife Conservation Commission Special Purpose Permit No. WX05089, Florida Department of Environmental Protection Research and Collection Permit No. 5-05-41, and University of Georgia Institutional Animal Care and Use Committee approval No. A2005-10044-0.

Abundance Estimation and Efficiency Comparison

We estimated abundance for the SRS using the design-based estimator described by Thompson (2002). For ACS, abundance was estimated with the modified Horvitz-Thompson estimator using initial intersection probabilities (Thompson and Seber 1996):

$$\hat{\mu} = \frac{1}{N} \sum_{i=1}^{N} \frac{y_i I_i^!}{\pi_i^!}$$

where y_i represents the y-values of the sample units in the final sample and $I_i^!$ receives a value of 1 (with a probability of $\pi_i^!$) if the i_{th} sampling unit is included in the sample and 0 if not. We used ratio of estimator variances (var[SRS]/var[ACS]) to evaluate efficiency of ACS using variance equations from Thompson and Seber (1996). With this ratio, a value >1 indicates greater efficiency using ACS. We set sample size of SRS equal to the final sample size (including edge units) for ACS (Thompson and Seber 1996).

Bias Correction

Although SRS and ACS are design-unbiased estimates, imperfect detection and animal mobility can introduce bias (Otis et al. 1978, Wilson and Anderson 1985). We estimated capture and recapture probabilities of the trappable population in program MARK using 8 closed-population models which allow for individual and combined effects of behavior, capture heterogeneity, and time (White and Burnham 1999). Due to low number of captures, all individuals were pooled for analysis. We used Akaike's Information Criterion for small sample sizes (AIC_c) to evaluate and select the most parsimonious models (Burnham and Anderson 2002). We used the model-averaged abundance estimate of the trappable population to estimate detection rate. We divided estimates of abundance from design-based estimators by estimated detection rate to correct for imperfect detection (Thompson and Seber 1994).

To account for potential bias in abundance estimates due to captures of transient woodrats, we radiotracked 10 individuals (5M:5F) on 8 sampling units and estimated proportion of individuals with nests occurring outside of sampling units. We selected individuals opportunistically with a minimum weight of 180 g for attachment of 9-g radiotransmitter collars (AVM Instrument Company, Colfax, California). Eight radiocollared individuals were located during the day twice each week between April and July 2005. Two individuals were located twice each week between September-November 2005. We located nests by homing and recorded each nest location with a hand-held GPS unit. If multiple nests were used by an individual, we defined primary nest sites as those utilized $\geq 80\%$ of the time. We used a GIS to determine if locations of primary nests occurred within or outside of the network on which the individual was captured. We calculated a maximum-likelihood estimate (Williams et al. 2002) of the proportion of individuals with primary nest sites occurring outside of the sampled area. We corrected design-based estimates of abundance for bias due to movement by subtracting the estimated percentage of individuals with nests occurring outside the sampled area.

Poststratification

Trapping data and telemetry data collected during a concurrent study suggested that density of rock and debris piles and presence of large trees in mature hammock were also important features influencing Key Largo woodrat abundance. Therefore, we used poststratification (Thompson 2002) to evaluate the effectiveness of two additional strata delineations in addition to the original delineation. When conducting grid sampling, we observed additional rock and debris piles that were unknown to refuge personnel and were not identified in our initial searches. Therefore, to ensure accuracy of the original stratum delineation for poststratification analysis, we conducted thorough searches of sampling units and recorded locations of additional rock and debris piles. Therefore, we delineated the first poststratification design strata the same as in the initial design (\leq 75 m or >75-m from rock or debris pile) but included a more thorough survey of rock and debris piles. In the second design, we delineated two strata based on density of rock and debris piles (≤ 3 or > 3) within a 75-m radius of grid center. In the third design, we combined the two rock and debris density strata with a third stratum that delineated areas having ≤ 3 debris piles as occurring in mature forest (indicating presence of large trees used as nest substrate) or otherwise based on a GIS layer developed from previous studies (Ross et al. 1995; McCleery et al. 2006a, 2006b).

We obtained the variance of the poststratified population estimate using the following equation (Thompson 2002) in which the first term is the variance estimate for stratified random sampling with proportional allocation with a second term added due to random samples sizes in each stratum (h):

$$var(\tilde{y}_{st}) = \frac{N - n}{nN} \sum_{h=1}^{L} \left(\frac{N_h}{N}\right) \sigma_h^2 + \frac{1}{n^2} \left(\frac{N - n}{N - 1}\right) \sum_{h=1}^{L} \left(\frac{N - N_h}{N}\right) \sigma_h^2$$

Efficiency of each design was estimated as the ratio of the original SRS estimator variance to the estimator variance for poststratification, where a value greater >1 indicates increased efficiency due to poststratification. Using the poststratified design with the lowest variance, we calculated samples size needed to estimate abundance within 20%–80% of actual abundance with $\alpha = 0.1$ (Thompson 2002).

Results

We detected 11 individuals on 7 sampling units with the initial stratified random sample (n=40). Sampling in 33 adaptive units resulted in an additional 22 captured individuals. Twenty-three of the 33 grids added adaptively did not result in additional captures. The best closed-population model for estimating abundance of the sampled population received 79% percent of the AIC weight and included the effects of behavior and heterogeneity in capture probability. The model-averaged estimate of abundance for the sampled population was 34.4 (SE = 2.32; Table 1) resulting in an estimated detection rate of 0.96 (95% CI 0.85-1.0). Two of the 10 individuals were found to have nest sites outside of the sampled area resulting in an estimated 0.20 (SE = 0.06) positive bias from movement. Total population size, corrected for imperfect detection and movement, was estimated at 321 (95% CI 13-629) for SRS and 323 (95% CI 0-652) for ACS. The ratio of variance(SRS)/variance(ACS) was 0.47 with sample size equal to the final sample size for ACS (n = 73).

We detected additional rock and debris piles with intensive

Table 1. Model-averaged estimates of mixture (n = 2 groups; p_i), capture (p) and recapture probabilities (c), and abundance (N) for 33 Key Largo woodrats captured on 17 sampling units, 7 random and 10 adaptive, for four consecutive days April-May 2005, Key Largo, Florida.

Parameter	Estimate	SE	95% LCI	95% UCI
Probability of mixture (pi)	0.201	0.089	0.078	0.428
Group 1, day 1 (p)	0.654	0.223	0.215	0.929
Group 1, day 2 (p)	0.659	0.231	0.206	0.935
Group 1, day 3 (p)	0.506	0.309	0.083	0.920
Group 1, day 4 (p)	0.693	0.260	0.171	0.961
Group 2, day 1 (p)	0.483	0.095	0.308	0.662
Group 2, day 2 (p)	0.445	0.127	0.226	0.687
Group 2, day 3 (p)	0.565	0.185	0.229	0.850
Group 2, day 4 (p)	0.582	0.207	0.208	0.881
Group 1, interval 1 (c)	0.040	0.096	0.000	0.852
Group 1, interval 2 (c)	0.040	0.096	0.000	0.850
Group 1, interval 3 (c)	0.071	0.127	0.002	0.769
Group 2, interval 1 (c)	0.879	0.071	0.664	0.964
Group 2, interval 2 (c)	0.894	0.077	0.633	0.976
Group 2, interval 3 (c)	0.851	0.078	0.630	0.950
Population size (N)	34.414	2.324	29.859	38.968

 Table 2. Comparison of abundance estimates (N) and estimator variance for Key Largo

 woodrats sampled April–June 2005, Key Largo, Florida, from stratified random sampling as

 originally defined, with strata (K) misclassified, versus three designs using post-stratification

 with refined strata delineations.

Design	К	N ^a	Variance	Efficiency ^b
Original SRS	2	386	23273.42	
Design 1 ^c	2	261	13832.38	1.68
Design 2 ^d	2	238	13520.37	1.72
Design 3 ^e	3	223	11077.97	2.10

a. Estimate of abundance not accounting for bias due to movement or imperfect detection

b. Efficiency measured as ratio of original SRS variance to post-stratified variance c. Two strata delineated as \leq 75 m or >75 m from rock or debris piles.

d. Two strata delineated as ≤ 3 or >3 debris piles.

e. Three strata delineated as >3 debris piles, ≤ 3 debris piles in mature hammock, and ≤ 3 debris piles in any other hammock age.

searching, resulting in six misclassified sampling units in the initial stratification design. Poststratification was an effective means of accounting for initial error in stratum delineations and resulted in lower estimator variances for all three designs examined. The third poststratification design, delineating three strata based on rock/debris pile density and forest age, was the most efficient (Table 2). The most precise estimate of abundance using poststratification, corrected for detection rate and movement, was 188 (95% CI 0–400). With this design, required sample sizes to estimate abundance within 20%–80% (α =0.10) of the actual value varied between 181–58, respectively (Figure 1).

Discussion

Our ACS estimator resulted in a greater number of individuals per sampling unit than SRS, but variance of the SRS estimator was lower making SRS a more efficient design for estimating Key Largo woodrat abundance. Efficiency of ACS over traditional designs, as measured by a decrease in estimator variance given equal cost, is dependent on both the degree of rarity and spatial distribution of individuals in the target population (Thompson and Seber 1996, Smith et al. 2004). To achieve a relatively low estimator variance with ACS the population must be rare and clustered, resulting in a high within-network and low between-network variance (Thompson and Seber 1996). In our study, 3/7 units (43%) that met the criterion for adaptive sampling did not result in additional detections, indicating that a portion of the population was not clustered. The zero within-network variance of these 3 units likely contributed to the high ACS estimator variance.

In addition to population distribution, sampling design efficiency can be strongly influenced by criteria for initiating adaptive sampling and the neighborhood definition, both of which affect final sample size (Brown 2003). Liberal criteria and neighborhood definitions can lead to a large final sample size, making ACS impractical for most monitoring situations. Final sample size can



Figure 1. Sample size (number of trapping grids) required to estimate Key Largo woodrat abundance within 20%–80% of the actual value ($\alpha = 0.10$) using a stratified random design, with three strata delineated by density of debris piles and forest age class. Sample sizes were calculated based on strata-specific sample variances from post-stratification of data collected on 40 trapping grids on north Key Largo, Florida.

be effectively limited with the use of stopping rules, which often biases estimates of abundance due to inaccurate estimates of network inclusion probabilities (Brown 1994, Lo et al. 1997, Brown and Manly 1998, Salehi and Seber 2002). However, a small amount of bias introduced with a stopping rule may be outweighed by the increase in efficiency gained through reduced sample size and cost (Lo et al. 1997). We applied a stopping rule of one adaptive addition to limit final sample size, but our final sample size was still relatively high which likely is attributable to the criteria for initiating adaptive sampling being too liberal. Increasing the criteria to 2 individuals would have resulted in only 1 less individual but 17 fewer sampling units, lowering the cost of the study (i.e., number of trapping grids required) by 23%.

Initial inaccurate strata delineations resulted in a high estimator variance, approaching that of a simple random sample. The effect of accurate strata delineations on estimator variance was demonstrated with poststratification, resulting in an approximately 40% decrease in variance. Poststratifying into three strata including areas with high density of available nest substrate, areas with low artificial substrate density but with presence of natural substrate, and all other areas gave the most precise abundance estimate, reducing the original SRS estimator variance by more than half. Mature forest was considered a valuable stratum delineation because Key Largo woodrats use the root systems and downed logs of large overstory trees found in mature forests as nest sites, although at a lower frequency than rock and debris piles (McCleery et al. 2006b). Utility of mature hammock as a stratum was supported by the fact that all individuals captured on sampling units >75 m from debris piles were in mature hammock.

We found that bias introduced by imperfect detection was minor (estimated 4% negative bias) likely because Key Largo woodrats exhibit high capture and recapture probabilities (i.e., highly trappable). Although bias due to imperfect detection was low in our study, estimating detection rate with marked animals requires little additional effort and should be incorporated into future abundance estimates as a precaution in case detection rate varies over time as the population increases or decreases. In contrast, individuals captured on our grids but with nests outside of our sampled units (transient individuals) resulted in considerable positive bias. Key Largo woodrats utilize multiple nests (female mean = 1.63 ± 0.16 nests, male mean = 2.25 ± 0.19 nests; Winchester 2006) within their home range. Furthermore, home range size is large enough (female mean = 0.21 ± 0.048 , male mean = 0.48 ± 0.12 ha; McCleery et al. 2006a) to incorporate ≥ 2 of our 0.56 sampling units if nests were located near the edge of the units. We found that 2/10 (20%) of radiotracked individuals had primary nests, which we defined as nests used \geq 80% of the time, outside of our sampled units. Accounting for potential bias associated with transient individuals requires considerable effort and cost if radiotracking is not a part of the original study design. Nonetheless, because of the possibility of overestimating the population, accounting for this bias is necessary to obtain reliable Key Largo woodrat population abundance estimates.

Management Implications

The best sampling design among those considered in our study was a SRS with three strata delineated by density of artificial substrate and forest age. However, the population estimate obtained using this design produced unacceptably wide confidence intervals (n=188; 95% CI 0-400) using our original 40 sampling units. Our sample size calculations suggest that 180 sampling units would be required to estimate abundance within 20% of the actual value with 90% confidence using this design. Therefore, estimating abundance of the Key Largo woodrat population is likely infeasible given the budgetary and personnel constraints of agencies responsible for monitoring the population. Depending on management goals, it may be more cost-effective to restrict monitoring to smaller spatial scales, particularly as habitat improvement is likely to be applied incrementally at small scales. Small-scale monitoring would provide essential feedback on the response to habitat improvement while greatly reducing the effort required to monitor the total population.

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