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Traditional methods for estimating white-tailed deer population size and density are affected by behavioral biases, poor detection in densely forested areas, and invalid techniques for estimating effective trapping area. We evaluated a noninvasive method of capture–recapture for white-tailed deer *Odocoileus virginianus* density estimation using DNA extracted from fecal pellets as an individual marker and for gender determination, coupled with a spatial detection function to estimate density (spatially explicit capture–recapture, SECR). We collected pellet groups from 11 to 22 January 2010 at randomly selected sites within a 1-km^2 area located on Arnold Air Force Base in Coffee and Franklin counties, Tennessee. We searched 703 10-m radius plots and collected 352 pellet-group samples from 197 plots over five two-day sampling intervals. Using only the freshest pellets we recorded 140 captures of 33 different animals (15M:18F). Male and female densities were 1.9 (SE = 0.8) and 3.8 (SE = 1.3) deer km^{-2}, or a total density of 5.8 deer km^{-2} (14.9 deer mile^{-2}). Population size was 20.8 (SE = 7.6) over a 360-ha area, and sex ratio was 1.0 M: 2.0 F (SE = 0.71). We found DNA sampling from pellet groups improved deer abundance, density and sex ratio estimates in contiguous landscapes which could be used to track responses to harvest or other management actions.

White-tailed deer *Odocoileus virginianus* population data are needed for scientific management decisions, particularly because harvest is an important management tool (reviewed by DeYoung 2011). Population estimates are needed to maximize recreational opportunity and keep deer populations within the bounds set by managers (Hamilton et al. 1995, Gibbs 2000). White-tailed deer are the most popular game animal in the USA, the number of days spent deer hunting and the associated expenditures exceeding that of all other species combined (US Fish and Wildlife Service et al. 2007). During the last 20 years increasing white-tailed deer populations have caused a rise in crop and property damage and deer–vehicle collisions (Conover et al. 1995, Romin and Bissonette 1996). High deer density also influences the structure and composition of forest understory (Tilghman 1989, Rossell et al. 2005) and chronic over-browsing can limit availability of food and cover for deer and other wildlife species (Casey and Hein 1983, deCalesta 1994) and can impact both faunal and floral species diversity (Anderson and Katz 1993, Rossell et al. 2005, 2007, Webster et al. 2005).

Population size (N) is a commonly estimated parameter for deer, but in contiguous environments, population boundaries are indiscreet and it can be difficult to determine the area to which the estimate applies. Population size for an abundant species such as white-tailed deer is a relevant parameter only when the area occupied by the sampled population is known and discrete (Parmenter et al. 2003). A variety of methods have been developed to estimate this area (A), from which density (D) can be estimated (N/A), ranging from boundary strips (Dice 1938) to radio telemetry to estimate the proportion of time animals spend within prescribed study area boundaries (Garshelis 1992). The former method can be biased high when movements are truncated at traps (Obbard et al. 2010), and the latter method requires radiomarking large numbers of animals. Some methods (e.g. track count, pellet-group count and direct counts on transects) produce widely varying estimates of density (Mandujano and Gallina 1995) and others (e.g. hunter observations, spotlight counts, infrared-triggered camera surveys, forward-looking infrared surveys, pellet-group counts) produce only a measure of relative abundance (Bennett et al. 1940, Rice and Harder 1977, McCullough 1982, Wiggers and Beckerman 1993, Jacobson et al. 1997, Belant and Seamans 2000, Koenen et al. 2002). Thus, deer managers need a population metric that can be used to quantify population responses to management activities that are spatially relevant.

Because of the difficulty in estimating the area sampled for applying traditional mark–recapture estimates, Efford (2004) developed a technique whereby D is estimated without defining an effective sampling area (A). Efford (2004) used a two-parameter spatial detection function to represent the capture process and fit a linear model. Animal range centers within the study area are directly correlated with the overall density of that area. Each animal is assumed to occupy a range center at an unknown location and each trap (detector) is set at a known location. Therefore, the probability of capture is a declining function of distance between the range center and the trap location, similar to the detection function in distance analysis (reviewed by Efford 2004). Although neither the individual range center nor the complete ranges of movement coordinates are fully observed, they can be predicted (Royle and Young 2008). This explicit capture–recapture method (SECR; Borchers and Efford 2008) produces density estimates unbiased by edge effects and incomplete detection (Efford et al. 2004). Moreover, the method can also produce estimates of population abundance (N) that are unbiased due to spatial heterogeneity (Efford and Fewster 2013), which reduces or eliminates a substantial source of potential bias that can plague non-spatial estimators.

Traditional capture–recapture techniques use physical markers (e.g. eartag, tattoo, branding) that require capture, handling and observing marked individuals. An alternative is to use genetic sampling to obtain capture–recapture data (Taberlet et al. 1997, Woods et al. 1999, Mowat and Strobeck 2000, Waits and Paetkau 2005). Genetic sampling ensures tag permanency, decreases intrusiveness and can increase capture probabilities, reduce capture bias and shorten the sampling period (Taberlet et al. 1999, Woods et al. 1999, Mills et al. 2000). Scat and hair are the most common sources of DNA obtained for genetic sampling (Miller et al. 2005). Feces contain cells shed from the intestinal lining and DNA extracted from those cells can be used as an individual marker. Thus, this technique can potentially provide abundant samples for capture–recapture studies (Maudet et al. 2004) at low cost (Foran et al. 1997, Brinkman et al. 2011), and without animal capture or disturbance (Belant 2003, Belant et al. 2005).

Population estimation using genetic sampling from feces was first successfully conducted for coyotes *Canis latrans* (Kohn et al. 1999) followed by studies on other carnivores (Ernest et al. 2000, Creel et al. 2003, Wilson et al. 2003, Flagstad et al. 2004, Hedmark et al. 2004, Bellemain et al. 2005). Although use of genetic material obtained from pellet groups has been used to census a variety of ungulates (Valière et al. 2007, Harris et al. 2010, Brinkman et al. 2011, Poole et al. 2011), fecal DNA has not been used to estimate population size of free-ranging white-tailed deer. Also, no one to our knowledge has used SECR for density analysis for fecal sampling on any ungulates.

We hypothesized that spatially explicit capture–recapture methods using genetic sampling may be a viable option for monitoring white-tailed deer herds. Our objective was to evaluate the effectiveness of genetic sampling from scat (pellet groups) to estimate white-tailed deer abundance and density using SECR techniques.

Study area

Arnold Air Force Base (AAFB) is approximately 112 km southeast of Nashville, positioned between the towns of Manchester, Tullahoma and Winchester, and is within the Duck and Elk river watersheds in Coffee and Franklin counties, Tennessee (US Dept of Defense 2006; Fig. 1). White-tailed deer are an important natural resource for hunting at AAFB, but also impact forested habitats and cause deer–vehicle collisions. The deer population on AAFB is managed jointly by Dept of Defense and the Tennessee Wildlife Resources Agency (TWRA). A majority of the area on AAFB is open to public hunting and is managed as a Wildlife Management Area by TWRA. AAFB is typical of many areas where deer are managed in that techniques used to estimate white-tailed deer populations are impaired by logistical problems and biases that culminate in loss of precision and accuracy.

AAFB was 15 815 ha in size with forest comprising 11 553 ha. Forest consisted of cultivated loblolly pine *Pinus taeda* plantations or hardwood forests dominated by oaks *Quercus* spp. with mid- and understories primarily consisting of dogwoods *Cornus* spp., maples *Acer* spp., sassafras *Sassafras albidum*, sourwood *Oxydendrum arboretum*, blueberries *Vaccinium* spp., hickories *Carya* spp. and blackgum *Nyssa sylvatica*. Grasslands and early-successional vegetation in utility rights-of-way occupied 898 ha. The remaining 1895 ha of the installation consisted of wildlife food plots, buildings and structures, mowed areas and other open areas (e.g. landfills, roads; US Dept of Defense 2006).

Our field sampling was concentrated within a 100-ha area in Unit 1 of AAFB (Fig. 1). Unit 1 consisted of 51.0 ha of hardwood, 38.3 ha of pines and 7.2 ha of fields. The hardwood sections were dominated by post oak *Quercus stellata* (35.7 ha) and southern red oak *Quercus falcata* (15.3 ha). The proportion of vegetation types was similar across all of AAFB. We developed a forest cover map for the study area by manually digitizing the area based on orthophotoquad maps provided by AAFB with imagery from Google Earth according to Basinger (2013). Mature deciduous closed-canopy forest cover was all classified as forest. Early to late age classes of planted pines were combined as pine. All other types were classified as open which included fields and road shoulders.

Material and methods

Survey design

We generated 150 random locations using Arc Map (ESRI ArcMap 9.3.1, 1999–2009) for field sampling during each of five sampling occasions (total of 750 GPS locations); each sampling occasion consisted of a sequential two-day period with no time between occasions. We added a 20-m radius buffer around each sample location to prevent sample plots from overlapping during individual capture occasions. To decrease the probability of mixing pellet groups from >1 individual, we collected only pellets adjacent to each other within the pellet group and excluded pellets scattered more than about 0.1 m from the center of the group (Harris et. al 2010). Fresh latex gloves were used to collect each sample.

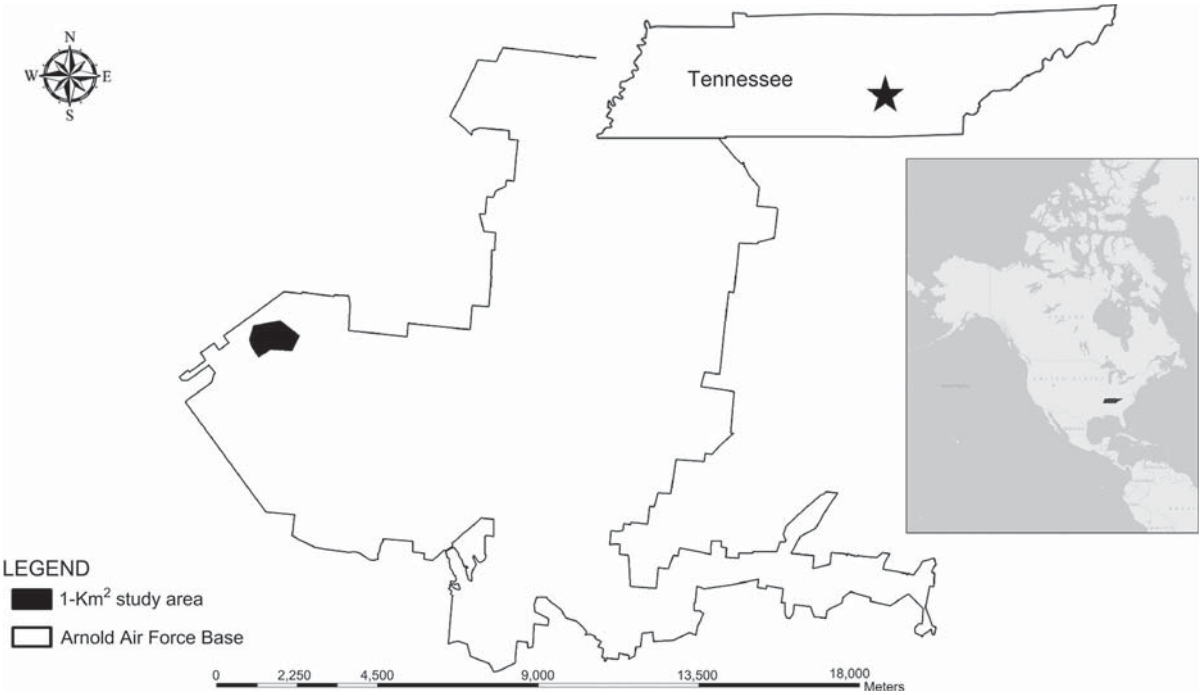

Figure 1. Arnold Air Force Base, Coffee and Franklin Counties Tennessee, USA for collection of white-tailed deer pellet groups, January 2010. The 1-km^2 sampling area is in black.

We rated samples for quality based on three categories: a sample coded as 1 was considered fresh and of high quality (still moist, intact, and above litter), 2 represented intermediate quality and freshness (somewhat moist, intact), and 3 represented low quality and older age (dry, easily broken, under litter; Brinkman et al. 2010). We placed fecal pellets in labeled paper bags. All pellet groups were removed from plots to avoid resampling in later sampling occasions. Samples were allowed to dry at room temperature and desiccant was placed around the paper bags in a storage box to facilitate drying (D. Paetkau, Wildlife Genetics International, pers. comm.).

Genotyping

Microsatellite analysis was conducted by Wildlife Genetics International (WGI). Fecal pellets were analyzed for individual genetic profiles and gender determination. Pellets that were broken, crushed or clumped were not analyzed because of the high probability that inhibitory secondary plant compounds within the feces would co-purify with the DNA and thus compromise success rates (Wilson 1997). WGI immersed 1–6 pellets, depending on size and availability, in Qiagen's ATL digest buffer in a test tube which was agitated by gentle swirling several times per h. Pellets were removed from the solution and DNA purification followed methods described in Woods et al. (1999) and Paetkau (2003). Standard cycling and buffer conditions for PCR were performed according to Paetkau et al. (1998). The PCR products were visualized by capillary electrophoresis on a DNA sequencer, and scored with the assistance of Genotyper 2.1 software.

WGI selected 21 microsatellite markers routinely used in parentage projects involving white-tailed deer to determine individual identification (Genbank accession no. or reference in parentheses): BL25 (Bishop et al. 1994), BL42 (Bishop et al. 1994), BM4107 (G18519), BM6438 (G18435), BM6506 (G18455), FCB193 (L01533), Inra11 (Bishop et al. 1994), OhemK (AF102242), OhemD (AF102247), OhemH (AF102254), OhemN (AF102244), OhemO (AF102245), OhemP (AF102240), OhemS (AF102258), OhemQ (AF102241), OhL (AF102257), OvirA (L35576), Rt5 (U90738), Rt7 (U90740), Rt13 (U90743), and Rt24(U90746). WGI excluded markers that amplified weakly from fecal samples, including most markers with allele lengths >200 bp, because amplification of DNA from fecal extracts was sensitive to the length of the DNA segment analyzed. However, the longest locus considered was BL42 (up to 278 bp long). After removing markers problematic for success rate (OheK, OheQ, OhL and Rt13), and sequencing sufficient samples to have >15 animals genotyped at each marker, WGI compared 17 markers for variability, success rate and ability to fit together efficiently in a single sequencer lane (Table 1). Finally, WGI selected six markers with high heterozygosity and ability to individually identify many individuals (BL42, BM4107, FCB193, OhemS, Rt5, Rt7; Table 1) and added a ZFX/ZFY gender marker alongside the microsatellites (D. Paetkau, Wildlife Genetics International, unpubl.). The ZFX/ZFY primers amplified a 204 bp Y peak and 193 bp X peak in deer and were designed explicitly because of the low success rate caused by excessive length of the published primers.

Genotyping errors can result in positively biased parameter estimates if samples from the same individual are assigned different genotypes. Conversely, if the markers are not sufficiently variable, too few individuals will be identified resulting in a negative bias (Woods et al. 1999, Mills et al.

Table 1. Microsatellite marker variability, observed and expected heterozygosity and observed number of alleles of white-tailed deer on Arnold Air Force Base, TN, USA, 2010. The first 6 markers, and a gender marker, were run on every sample for the purpose of individual identification.

Locus	N[a]	He[b]	Ho[c]	A[d]
BM4107	39	0.81	0.75	8
BL42	39	0.83	0.75	8
FCB193	39	0.79	0.8	8
OhemS	39	0.84	0.9	12
Rt5	39	0.9	0.9	8
RT7	39	0.78	0.78	10
6-locus mean		0.82	0.81	9
OhemN	18	0.8	0.94	6
OhemP	17	0.75	0.77	6
OvirA	25	0.79	0.88	6
OhemH	28	0.73	0.71	5
Rt24	19	0.67	0.79	4
BL25	15	0.33	0.4	2
BM6438	16	0.8	0.94	6
BM6506	23	0.78	0.74	7
Inra11	33	0.69	0.7	5
OhemO	18	0.16	0.06	3
OhemD	31	0.49	0.48	5
17-locus mean		0.70	0.72	6.4

[a]no. of white-tailed deer identified using given locus
[b]expected heterozygosity
[c]observed heterozygosity
[d]no. of observed alleles

2000). WGI used a combination of objective (peak height) and subjective (appearance) criteria to classify genotype scores. Samples were stratified into three groups: those that yielded high-confidence scores for 6 or 7 markers on the first pass, those that yielded high-confidence scores for 3–5 markers, and those that yielded ≤ 2-locus genotypes. Those that yielded high-confidence scores for 3–5 markers during the first pass were re-analyzed three more times at all 7 markers to evaluate run-to-run data reproducibility. A multilocus consensus genotype was then identified for each of those samples, and each repeat was assigned a value of 0, 0.5 or 1 based on whether 0, 1 or 2 alleles, respectively, matched those in the consensus genotype. These values were averaged across all 4 repeats of all 7 markers to arrive at q, the likelihood that a genotype belonged to a unique individual. Samples with $q < 0.6$ were culled based on prior experience. Lower values typically did not have enough DNA to verify any inconsistencies. Samples with $q > 0.6$, whereby the four repeats of analysis did not produce at least two high-confidence scores (identical to the consensus genotype for a given marker), were subjected to further rounds of single-locus analysis to confirm the genotype. Samples with ≤ 2-locus genotypes were culled. Finally, WGI re-analyzed markers that mismatched at 1 or 2 of 7 markers to check for errors, which was similar to the procedure described by Kendall et al. (2009). WGI found and corrected errors in 14 samples because of amplification mistakes at the OhemS and BL42 marker. These data checking protocols ensured that the number of individuals identified in the dataset was not inflated through undetected genotyping error (Kendall et al. 2008).

Abundance and density estimation

We used capture histories and capture locations of individuals identified based on their genetic profile to estimate white-tailed deer abundance and density using Program DENSITY (ver. 4.4; <www.otago.ac.nz/density/>, accessed 15 July 2011) and R package secr (ver. 2.5.0; <www.otago.ac.nz/density/SECRinR.html>, accessed 11 September 2013). Spatially explicit calculation of density involves a parameter for detection probability (g_0) and spatial scale (σ; Efford 2004). We modeled detection with the half-normal function because we assumed the distance from range center to zero capture probability was continuous and not a step function (Borchers and Efford 2008). We modeled the number of captured individuals as a homogenous Poisson distribution which is based on the assumption that animals were distributed randomly across the landscape (Borchers and Efford 2008). We considered that assumption to be reasonable because deer are not territorial or highly gregarious even though groups may temporarily cluster (e.g. does and yearlings or bachelor groups; DeYoung and Miller 2011). We modeled males and females separately whereby density (D) differed by habitat type (habtype) or was homogenous (1). We modeled the capture probability parameter (g_0) as a function of session time period (t), time before and after a rain event (17 January 2010) that occurred between sampling periods 3 and 4 (rain), as a two-class mixture for individual heterogeneity to delineate individuals into unobserved subgroups (h2), and as a constant (1). We also fit models with a behavioral response (b; because removing scats sampled in plots might affect subsequent recapture rates in plots that were resampled) on g_0. Spatial scale (σ) was modeled as a function of time period (t), with rain effects (rain), two-class mixture for individual heterogeneity (h2), and as a constant (1). Finally, we developed models with interactions of the most supported effects. We used an initial buffer width of 1500 m, which was based on multiplying root-pooled spatial variance (RPSV = 339.1 from initial analysis) × 4 as recommended by Efford et al. (2004). This was later increased to 2100 m because estimates for males were more stable with a slightly larger buffer. Estimates for σ based on a two-mixture heterogeneity model produced erratic estimates for males and those models were discarded. We compared models using Akaike's information criterion adjusted for small sample sizes (AIC_c; Hurvich and Tsai 1989). Models with lower AIC_c values were considered to have more support and be more parsimonious (Burnham and Anderson 2002). We compared differences in AIC_c values (ΔAIC_c) to evaluate the relative importance of the models; when models had a ΔAIC_c value of < 2, they were considered to have equal support. Model weights (w_i) were used to compare support and enabled us to make inferences about precision over the entire suite of models (Burnham and Anderson 2002). We used model averaging to calculate parameter estimates but used the 'derived' command in secr to obtain density estimates by gender. Finally we used the delta method to calculate the standard error for the sex ratio estimate (Powell 2007). To estimate population size, we first had to define a mask area over which the density estimate was to be applied. The size of that area is not critical but a mask area is required for the calculations. We arbitrarily assigned a 500-m buffer around

each plot which we thought was a biologically reasonable depiction of the relevant study population.

To assess whether our trapping grid was large enough, we simulated 10 sets of capture data given a trapping grid (12 × 12 m separated by 90 m and buffer of 2100 m, g_0 and σ as constant) and our estimates of D, g_0 and σ. We also doubled σ with other variables remaining unchanged.

Results

Field sampling

We collected pellet groups during 11–22 January 2010. Technicians searched 135–147 plots during each two-day sampling event which totaled 703 plots, 197 of which had ≥ one pellet-group (28%), resulting in 352 pellet-group samples (Table 2). The greatest number of pellet groups in one plot was eight (0.05%). Overall, 15% of the plots had one, 8% had two, and 3% had three pellet groups/plot. We searched 4% of the total area (140 10-m radius plots) per capture event, which yielded an average of 70 samples per capture event. A major rain event occurred on 17 January, between capture events 3 and 4. Mean number of pellet groups/sampling occasion prior to the rain event was 78 and mean number after the rain event was 58.

Genotyping

We used all 352 pellet-group samples for microsatellite analysis, 15 of which (4%) were inadequate for analysis. Of the 337 samples analyzed, 2 showed evidence of > 2 alleles per marker, suggesting a mixture of DNA from 2 individuals, and were not used. During error checking, genotypes that mismatched at 1 or 2 of the 7 markers ('1MM' and '2MM' pairs) were reanalyzed. Fourteen samples had errors that were corrected which included 8 amplification errors at OhemS and BL42. After correcting the errors, there were no remaining 1MM pairs and the 2MM pairs had been verified with reanalysis. One hundred and fourteen samples (34%) failed during genetic analysis and 223 samples (66%) provided individual identifications.

Percent success of genetic analysis varied by sample quality rating (79% for 1, 67% for 2, and 59% for 3). Samples noted as moldy, weathered, or falling apart during extraction generally failed analysis. Consequently, we used only fresh fecal pellets (rating 1) for subsequent analyses. Genotyping success rates also varied by collection date and sample abundance. Seventy nine percent of the total samples collected before the rain event (before 17 January) produced complete genotypes, compared with 28% after the event (included samples from all quality ratings). Samples comprised of a large number of pellets had a 75% success rate compared with 32% for samples with few pellets. Fewer pellets limited the number of high-quality pellets that could be used for extraction and reduced opportunities to reanalyze if errors occurred.

Table 2. Number of plots searched, number of plots with ≥ 1 sample (all pellet quality ratings), total number of samples for capture events, and average number of samples for white-tailed deer pellet groups, Arnold Air Force Base, TN, USA, January 2010. A rain event occurred between sampling period 3 and 4 (17 January 2010).

Sampling period	Plots[a]	Plots with pellet groups[b]	Total no. of samples[c]	Mean
1	135	47	95	
2	147	43	69	78[d]
3	139	38	72	
4	141	44	67	58[e]
5	141	26	49	
Total	703	198	352	

[a]no. of plots searched
[b]no. of plots with at least one sample
[c]total number of samples collected
[d]mean no. of samples collected before rain event
[e]mean no. of samples collected after rain event

Density estimation

We identified 33 individuals (15 males and 18 females) from 140 captures of the quality code 1 pellets. The mean maximum distance moved (MMDM) was 534.3 ± 64.1 m. The asymptotic range length was 978.7 ± 145.1 m. The mean distance between captures was 419.6 ± 24.1 m and the root pooled spatial variance was 345 m (simple measure of home range size).

The model for males that received most support included no habitat covariate effect on D, g_0 as a constant, and σ as a function of the rain event between weeks 3 and 4 (Table 3). The top model contained 46.3% of the weight but other models received some support so we model averaged to obtain parameter estimates. The estimate of σ for males was 527.0 m (SE = 102.2) and g_0 was 0.049 (SE = 0.013). Male density was 1.9 deer km^{-2} (SE = 0.8). For females, the top model included no habitat covariate effects on D, g_0 differed according to a rain event between weeks 3 and 4, and σ was constant, and contained 59.7% of the weight (Table 4). The estimate of σ for females was 376.9 m (SE = 62.0) and g_0 was 0.059 (SE = 0.016). Density was 3.8 female deer km^{-2} (SE = 1.3). Total deer density was 5.8 deer km^{-2} (SE = 2.1, 14.9 deer mile^{-2}). The sex ratio was 1.0 M: 2.0 F (SE = 0.71). The population size was 20.8 deer (SE = 7.6) over an area of 360 ha (the mask area after applying a 500-m buffer surrounding pellet collection sites).

We simulated 10 sets of capture data given a trapping grid (12 × 12 m separated by 90 m and buffer of 2100 m, g_0 and σ as constant) and our estimates of D, g_0, and σ. Estimates of D for males ranged from 1.22 to 3.28 ($\bar{x}$ = 2.26, SE among runs = 0.20, true D = 1.93), g_0 ranged from 0.033 to 0.061 ($\bar{x}$ = 0.047, SE among runs = 0.002, true g_0 = 0.048), and σ ranged from 397.2 to 548.9 ($\bar{x}$ = 485.5, SE among runs = 15.2, true σ = 508). Estimates of D for females ranged from 2.23 to 4.57 ($\bar{x}$ = 3.24, SE among runs = 0.26, true D = 3.88), g_0 ranged from 0.037 to 0.051 ($\bar{x}$ = 0.041, SE among runs = 0.002, true g_0 = 0.039), and σ ranged from 300.3 to 459.8 ($\bar{x}$ = 397.1, SE among runs = 13.4, true σ = 371). When we also doubled σ with other variables remaining unchanged our estimates of D for males were 2.21 (SE among runs = 0.1), g_0 was 0.048, SE among runs = 0.002), and σ was 930.9, SE among runs = 22.2). Likewise, estimates of D for females were 3.83 (SE among runs = 1.9), g_0 was 0.039, SE among runs = 0.001), and σ was 750.0, SE among runs = 21.0).

Table 3. Selected models for male white-tailed deer population estimation on Arnold Air Force Base, TN, USA, 2010. The half-normal detection function was used. Density (*D*) was modeled by habitat type (habtype) or was homogenous (1). Capture probability (g_0) and spatial scale (σ) parameters were modeled as a function of session time period (*t*), as time before and after a rain event between sampling periods 3 and 4 (rain), as having two-mixture individual capture heterogeneity (h2, pmix), and as a constant (1). We also fit models with a behavioral response (b) on g_0.

Model	No. of parameters	Log likelihood	AICc	ΔAICc[a]	AIC weight
D~1 g_0~1 σ~rain	4	–325.0721	662.144	0.000	0.4625
D~1 g_0~1 σ~1	3	–327.4976	663.177	1.033	0.2759
D~1 g_0~ rain σ~rain	5	–324.0957	664.858	2.714	0.1191
D~1 g_0~ rain σ~1	4	–326.7177	665.435	3.291	0.0892
D~1 g_0~b σ~rain	5	–324.9784	666.623	4.479	0.0493
D~1 g_0~h2 σ~1 pmix~h2	5	–327.4908	671.648	9.504	0.0040
D~1 g_0~1 σ~*t*	6	–324.8508	672.202	10.058	0.0000

[a]Akaike's information criterion adjusted for small *n*

Discussion

Our estimates of deer density (5.8 deer km^{-2}) were similar to values from Beaver et al. (2014; 4.0–6.6 deer km^{-2}) using aerial vertical-looking infrared imagery for a security area of AAFB where hunting was not allowed. However, using road-based ground surveys, Beaver et al. (2014) found densities were 3.0–7.6 times greater. Deer density was assumed to be higher in the security area than the management units of AAFB. We do not know the actual density in Unit 1, but our estimates using DNA from fecal pellets and SECR provided precise estimates of total population and also by gender. Therefore genetic sampling is an effective and precise method for density and estimation of sex ratio. This approach could easily be adapted to a larger area with different habitat types, using a stratified random sampling scheme and or habitat types as covariates (Morrison et al. 2001).

Genetic sampling and SECR from pellet groups allowed us to estimate density without the need to define trapping area boundaries. Also, estimates of population abundance are less biased because spatial heterogeneity is accounted for with spatially explicit models. For example, in traditional capture–recapture, animals whose home ranges occur along the boundary of the trapping grid may have lower capture probabilities than those within the grid, resulting in individual trap heterogeneity and negatively biased estimates. Gaps in the sampling grid can likewise result in unequal capture probabilities among individuals. Furthermore, spatially explicit models account for this heterogeneity when estimating *N* (Efford and Fewster 2013). Using SECR avoids the problems of determining an effective trapping area and so can easily be used for intensive data at rigorously collected subsamples to provide inference of density for a large region (Efford and Fewster 2013).

Most traditional population estimation techniques are unable to directly measure sex ratio and those that can assume similar detection and social behavior between genders (Downing et al. 1997, Jacobson et al. 1997, McCoy et al. 2011). Fecal genetic sampling provides individual identification and gender. Therefore, the techniques developed by Efford (2004) to model *D* can be used to determine unbiased sex ratios as well. Density can be derived by gender and an estimate of sex ratio and its uncertainty can be made. Techniques such as sighting or camera surveys to calculate sex ratios of white-tailed deer can be biased because they assume similar behavior for males and females, and estimates of female abundance are directly tied to estimates of male abundance (Downing et al. 1997, Jacobson et al. 1997, McKinley et al. 2006, McCoy et al. 2011). Even if estimates of *N* are obtained by traditional methods for each sex, the area effectively sampled for the two genders may not be equal and, consequently, the sex ratio estimate can be unreliable.

Closed capture–recapture methods assume the population is geographically and demographically closed. That

Table 4. Selected models for female white-tailed deer population estimation on Arnold Air Force Base, TN, USA, 2010. The half-normal detection function was used. Density (*D*) was modeled by habitat type (habtype) or was homogenous (1). Capture probability (g_0) and spatial scale (σ) parameters were modeled as a function of session time period (*t*), as time before and after a rain event between sampling periods 3 and 4 (rain), as having two-mixture individual capture heterogeneity (h2, pmix), and as a constant (1). We also fit models with a behavioral response (b) on g_0.

Model	No. of parameters	Log likelihood	AICc	ΔAICc[a]	AIC weight
D~1, g_0~rain, σ~1	4	–378.2510	767.579	0.000	0.5965
D~1, g_0~rain + b, σ~1	5	–377.2301	769.460	1.881	0.2329
D~1, g_0~ rain, σ~*t*	5	–378.2357	771.471	3.892	0.0852
D~1, g_0~*t*, σ~1	7	–374.3609	773.922	6.343	0.0250
D~1, g_0~ rain + b, σ~rain	6	–377.2156	774.068	6.489	0.0233
D~1, g_0~1, σ~rain	4	–381.8029	774.683	7.104	0.0171
D~1, g_0~1, σ~rain + h2, pmix~h2	6	–377.7627	775.162	7.583	0.0135
D~1, g_0~ rain + h2, σ~1, pmix~h2	6	–378.4857	776.608	9.029	0.0065
D~1, g_0~1, σ~rain × h2, pmix~h2	7	–376.8115	778.823	11.244	0.0000

[a]Akaike's information criterion adjusted for small *n*

assumption could be violated if pellet-groups remained viable for extended periods of time or if samples were collected during or immediately after the hunting season. However, after seven days of exposure to an environment with rain, Brinkman et al. (2010) was not able to genotype fecal pellets from black-tailed deer *Odocoileus hemionus sitkensis* because of degradation. Pellet groups that appeared older based on our rating system were less successful in genetic analysis than those rated as fresh. Thus, the general appearance of pellet groups was probably successful in eliminating old from newer pellets and reduce biases associated with population closure. Also, the ability to correctly identify pellet groups that are more likely to be successful for genetic analysis will reduce analysis costs.

Plots were randomly placed throughout the study area to facilitate equal detectability. Efford et al. (2009) found with SECR their results were unbiased even when animals were clustered, which could have occurred because of family groups (does and yearlings), bachelor groups, or commonly used areas and trails used by the population. Stratification of sampling area may increase success rate of finding samples and decrease time spent looking for samples. However, Langdon (2001) found that stratification based on cover type for pellet-group counts was unnecessary and would not have affected the success rate of pellet group collection. We detected an effect of the rain event on g_0 for females and on σ for males. The rain likely caused deterioration of pellets and reduced detection rates for females. For males, σ decreased after the rain, possibly as a result of older pellets (representing movements over a longer time period), deteriorating faster than more recent pellets. The possibility exists that deer may have left the study area over time as a result of our presence. However, we feel this is unlikely because telemetry studies on deer have shown that they quickly return to core areas after displacement by hunters and dogs (D'Angelo et al. 2003), and human activity is common in the area due to military training exercises, a managed hunt and vehicle traffic. Furthermore, as long as sampled deer left the study area at the same rate as unsampled deer, our density estimates would be accurate for the population on the area just prior to the first sampling occasion.

We intensively sampled a small area relative to average annual home-range size of white-tailed deer to ensure enough samples were collected to properly evaluate the technique. However, sampling areas that are too small can result in restricted detections of long-range movements and, thus, underestimates of σ and overestimates of density. Our sampling period was <2 weeks during winter when movements and home-range sizes were more restricted. The mean maximum distance of travel measured from minimum convex polygon home ranges of six GPS collared deer (4 male, 2 female; Basinger 2011) on AAFB during the sampling period was 1538.1 m (994.4–2320.8 m). Sollmann et al. (2012) found that SECR models were able to handle a wide range of trap spatial configurations and animal movements as long as the extent of the trap array was as big as or larger than the extent of individual movements during the study period. When we simulated 10 sets of capture data given a trapping grid (12×12 m separated by 90 m and buffer of 2100 m, g_0 and σ as constant) and our estimates of D, g_0, we found similar estimates to the true estimates from the simulated data. The similar results suggest that our trapping grid was indeed large enough to provide unbiased estimates, although variability was higher for males than females. We also found our grid was robust to moderate increases in σ.

Sampling could easily be conducted over a larger area using less intensive pellet group collection. Goode (2011) repeated the SECR analysis using 50% and 20% of the total pellet groups with individual genotypes. The 50% sampling scheme provided similar density estimates and precision to the total sample so we feel confident fewer samples in larger areas would yield precise estimates in many situations. If habitat types differed greatly, a stratified sampling approach could be conducted to assess the potential for density to differ across specified areas.

Genetic sampling is costly compared with other techniques used for estimation of white-tailed deer density. Five to six technicians collected pellet groups 8 h day^{-1} for 10 days. Total cost of collection materials (e.g. latex gloves, paper bags, pens) and storage materials (desiccant) was low ($<$\$100). Genetic analysis costs were \$60 for each sample analyzed, regardless of the number of runs for error checking (\$50 for genetic profile and \$10 for gender determination). We spent approximately \$20 000 for genetic analysis; however, our results indicated that we could have used half the samples to provide density estimates and measures of precision similar to using all samples. Beaver et al. (2014) estimated \$10 000 for one aerial survey on AAFB and considered the less costly road-based distance sampling to be of limited use because of the violations of random sampling, need for large sample size, and biased estimates. Therefore, genetic sampling with SECR may be an efficient method for unbiased estimates with high precision and the ability to determine sex ratios.

Additional information can be obtained from pellet groups including genetic diversity, habitat use, forage preference, movements, stress hormone levels and disease (Robbins et al. 1975, Collins and Urness 1981, Millspaugh et al. 2002, Millspaugh and Washburn 2004, Harris et al. 2010). Thus, pellet group collection and genotyping might be better justified if multiple hypotheses about deer biology could be simultaneously tested. The ability to associate individuals (genotyping) and individual characteristics (gender) with other information gained from pellet groups would improve our understanding of deer biology.

Conclusions

Genetic sampling using fecal pellets was effective to obtain precise estimates of density and sex ratio of white-tailed deer in a forested landscape. Our study indicates genetic sampling can be an important tool for white-tailed deer management because it allows managers to sample regardless of season and vegetation cover, where other techniques may not be applicable. Genetic sampling combined with SECR is able to account for problems associated with effective trapping area (Sollmann et al. 2012), and provide unbiased sex ratio estimates important for white-tailed deer management. Additionally, annual samples on an area may allow estimates of survival and fecundity using models developed by Pradel (1996). Genetic sampling is becoming increasingly available and is a reliable alternative to other density estimation

techniques for free-ranging ungulates that are rare, reclusive, difficult to capture, or live in dense cover that makes observation difficult.

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