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Population Genetics and Spatial Ecology of Bobcats (*Lynx rufus*) in a Landscape with a High Density of Humans in New England

Amy E. Mayer^{1,*}, Thomas J. McGreevy Jr.¹, Mary E. Sullivan², Charles Brown³, Thomas P. Husband¹, and Brian D. Gerber¹

Abstract - Lynx rufus (Bobcat) is a wide-ranging and highly adaptable predator whose populations are increasing throughout much of its natural range including in the New England states, yet there are only limited empirical ecological studies there. How Bobcats are responding to the unique modern landscape of southern New England with its highly forested landscape coupled with high density of humans is unknown. This lack of spatial and population ecological information impedes evaluating recovery and management objectives and identifying necessary management actions. Our objectives were to better understand the spatial and population structure of Bobcats in Rhode Island. We specifically examined space use, resource selection, and population genetics. We trapped Bobcats across 5 field seasons from April 2015 to March 2019, totaling 2232 trap nights. We captured 8 Bobcats, equipped GPS collars to a subset (n = 3), and collected locations for 4 to 9 months. We used GPS locations to estimate annual and seasonal home-range size and individual-level seasonal resource selection within the home range for each individual. Further, we used tissue samples collected from trapped individuals and opportunistically collected roadkill (n = 30)to examine the population genetic structure and effective population size of Bobcats in the state. We found the mean winter and summer home-range sizes were 219.3 km² and 51.7 km², respectively. Bobcats selected for forested wetland habitats and were associated with areas closer to wetlands and young forests, according to resource-selection models. They also selected for areas with higher road densities, yet avoided developed areas. We found that Bobcats in Rhode Island are part of 1 genetic population and estimated their effective population size to be 82 individuals (95% CI: 44-329). Our study highlights the importance of examining a widely distributed species at a local scale in order to employ evidence-based management practices.

Introduction

Lynx rufus (Schreber) (Bobcat) is the most widely distributed felid in North America. The species is a generalist predator that uses a wide variety of land-cover types, including boreal and temperate forests, coastal swamps, bottomland hard-woods, and deserts (Kelly et al. 2016). The diet, body size, pelt appearance, and home-range size of Bobcats vary considerably throughout their range. As an apex or mesopredator and an important harvested furbearer species, the management of

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Bobcats is of particular importance to wildlife managers; information on their distribution, abundance, and habitat use are commonly cited as research needs (Bluett et al. 2001). However, their cryptic behavior and low density can make them a difficult species to study (Gommper et al. 2006, Long et al. 2007). State wildlife agencies often rely solely on public sightings, hunter surveys, or harvest data to estimate population status and trends (Anderson and Lovallo 2003, Roberts and Crimmins 2010). Based on these survey methods, 32 states in the contiguous United States have reported that populations are increasing, and population estimates in the US overall have more than doubled since 1982 (Roberts and Crimmins 2010). Because public sightings and hunter surveys on their own often do not accurately reflect a species' abundance (Mahard et al. 2016), the actual extent of these reported population changes are largely unknown. However, they are likely a result of changes in land-use practices, harvest regulation, and natural range expansion (Roberts and Crimmins 2010).

Three of the 6 New England states (i.e., New Hampshire, Vermont, and Connecticut) report increasing Bobcat populations, with only Vermont conducting a harvest analysis to estimate population status, while Connecticut and New Hampshire use public sightings, vehicle collisions, or incidental harvest numbers to estimate their population status (Roberts and Crimmins 2010). In recent years, Bobcat populations have been studied more intensively in the region, particularly in northern New England, with studies on habitat selection and use (Abouelezz et al. 2018, Broman et al. 2014, Reed et al. 2017), occurrence (Long et al. 2011), home-range requirements (Donovan et al. 2011), and population structure (Carroll et al. 2019). Bobcat populations in southern New England states have received less attention, resulting in a lack of basic ecological information for wildlife agencies to use in management and decision-making. In Rhode Island, the number and distribution of unsolicited public sightings and road kills compiled by the Division of Fish and Wildlife at the Rhode Department of Environmental Management (RIDEM) over the past 20 years has increased from an average of 5.4 Bobcat sightings per year between 1999 and 2003, to an average of 11.4 per year between 2011 and 2015 (C. Brown, unpubl. data), which is consistent with reported region-wide trends, yet the status of the population is unknown.

Historically, Bobcat populations experienced marked reductions throughout their range due to unregulated harvest prior to their inclusion in Appendix II of the Convention on International Trade in Endangered Species, predator-control practices such as federal trapping programs (Crowe 1975) and state-paid bounties (Litvaitis et al. 2006), and competition from expanding *Canis latrans* Say (Coyote) populations (Litvaitis and Harrison 1989, Robinson and Grand 1958). While Bobcats were never considered extirpated from Rhode Island, they were classified as "rare" in 1968 by state biologists (Cronan and Brooks 1968), and more recently were classified as state threatened and a species of greatest conservation need under the 2015 State Wildlife Action Plan (Rhode Island Department of Environmental Management 2015). Rhode Island is the second most densely populated state in the US, with an average of 263 people per km² (US Census Bureau 2018).

Yet, there is a high proportion of mature forest cover in the state, such that Bobcats are occupying this unique landscape that is highly forested and has a large anthropogenic footprint. As a species with relatively large home-range requirements, Bobcats are vulnerable to habitat fragmentation and loss caused by human development (Crooks et al. 2010, Long et al. 2011). In Rhode Island, important Bobcat habitat, such as early successional forest and shrubland, currently cover only 3.3% of the state's total land area and is currently declining (Buffum et al. 2011). The current distribution and space-use of Bobcats in the state is unknown, and despite the need for this information to guide management, there has yet to be any empirical studies on Bobcat in the state of Rhode Island. Two important aspects of an animal's ecology are their physical movements and the movement of the animal's genes in their environment.

Understanding the population structure of a species is an important factor in making informed management decisions (Coltman 2008). This knowledge allows managers to track an animal's gene flow through the environment and evaluate how differential harvest on subsets of the population (e.g., sex, age) may affect the effective population size and hence genetic diversity of the population. Recently, an extensive population structure analysis of Bobcats in northern New England and Canada identified genetic structure that was likely historically influenced by major barriers to their gene flow, such as the White Mountains (Carroll et al. 2019). The human population density in Rhode Island is high, and the state has an extensive road network, which could impede dispersal and thus gene flow.

Understanding the home-range requirements of any species also can be important for making informed management decisions (Donovan et al. 2011). A species' home range provides insights into the total area needed for individuals to acquire the necessary resources to survive and reproduce (Burt 1943). Furthermore, changing space-use patterns may be a warning sign for population changes, such as reduced resources leading to animals moving further to obtain necessary resources, which may lead to higher mortality risk (e.g., due to vehicle collisions). The home-range area of Bobcats is known to be highly variable due to both demographic and ecological factors. Home-range area and space-use patterns can vary based on sex and climatic season (Litvaitis et al. 1987, Plowman et al. 2006) as well as availability of prey and other resources (Litvaitis et al. 1986). Additionally, Bobcat home-range sizes have been shown to vary depending on the population's geographic location, with home-range estimates varying from 2.6 km² in the southern portions of their range (Miller and Speake 1979) to as large as 112.2 km² in the northern-most parts of their range (Litvaitis et al. 1986).

Within the home range, identifying and characterizing resource requirements are essential in guiding land-use management practices aimed at protecting critical habitat. Resource-selection functions (RSF) are used to identify resources and habitat types that are selected for by individuals and populations within a specific area (Manly et al. 2002). Several studies on Bobcats have documented habitat preferences in the northern portion of their range (Broman et al. 2014, Reed et al. 2017), as well as in southern coastal areas (Roberts et al. 2010) and

the Midwest US (Tucker et al. 2008). However, a lack of data exists in southern New England, particularly in Rhode Island, where the temperature, topography, and available habitats differ from the prior studies examining Bobcat resource selection. The percentage of land area covered by forested wetlands and swamps (9.7%) and coastal shrublands (1.7%) is higher in Rhode Island than in northern New England states, and the species composition of forested areas differs from the northern states (Anderson et al. 2013). Additionally, features such as rugged, rocky outcrops and ledges or areas of high slope often identified as preferred habitats of Bobcats (Broman et al. 2014, Donovan et al. 2011, Reed et al. 2017) are largely absent from the Rhode Island landscape. These differences in resource availability make it important to characterize Bobcat resource selection within the state of Rhode Island to guide population management and land conservation decision-making. Additionally, documenting Bobcat resource use in an area already heavily influenced by human development will be useful for other states in the region to plan for future management of the species as impacts from development and fragmentation continue to alter the landscape.

In this study, our objectives were to better understand the spatial and population ecology of Bobcats in a forested landscape with a high density of humans by: (1) estimating the population-level genetic structure of Bobcats in Rhode Island, (2) estimating the size of home-range and core-use areas on an annual and seasonal basis, and (3) estimating seasonal resource selection for each individual. We expect that Bobcats in Rhode Island represent a single genetic population, given their potential for large-scale movements, relative to the size of the state. We predicted that Bobcat home ranges in Rhode Island would be larger than those reported in recent home-range estimates in northern New England due to higher human densities and more fragmented landscapes in our study area. Also, we predicted that forested wetlands would be important resources due to their prevalence on the landscape, and that resources associated with anthropogenic land use would be used more frequently than in prior studies due to more availability of these features.

Methods

Study area

Our study took place from the spring of 2015 to summer of 2019 in Rhode Island. We opportunistically collected genetic data statewide; however, our Bobcat trapping efforts were limited to Washington County, RI, where we have had a higher number of Bobcat captured on field cameras (A.E. Mayer, unpubl. data). Dominant vegetation types in the study area were *Quercus* (oak) and mixed deciduous forests, and forested wetlands. Monthly average temperatures during the study varied from 3 °C to 16 °C in the winter months (January to April, or leaf-off period) and 17 °C to 30 °C in summer months (May to October, or leaf-on period), with average monthly precipitation of 11 cm during the winter and 9 cm during the summer (Weather Underground 2019).

Trapping and animal handling

We trapped Bobcats over 5 field seasons (March–April 2015, October 2015– April 2016, October 2016–May 2017, November 2017– June 2018, January–March 2019) focused on the southern coast of Rhode Island (Washington County). We selected trap sites based on sighting reports from the public, scouting with trail cameras, and examination of satellite imagery to identify natural or man-made features that might direct Bobcat movement through a particular area. We used 3 types of cage traps: Tomahawk Model 209.5 (Tomahawk Live Trap Co., Tomahawk, WI), Camtrip Cages (Barstow, CA), and Comstock double-door traps (Comstock Custom Cage, LLC, Gansevoort, NY) baited with commercially available scent lures, visual attractants (such as feathers), and food baits (e.g., rabbits, squirrels, and ducks) obtained as road kill or donated by hunters and trappers.

We immobilized captured Bobcats using a mixture of 10 mg/kg ketamine and 2 mg/kg xylazine through intramuscular injection using a jab-stick. We collected morphometric measurements, determined the sex, and aged animals (adult or juvenile) by evaluating dental eruption (Jackson et al. 1988). For adults, we removed a front lateral incisor for aging using cementum annuli analysis (Matson's Laboratory, Milltown, MT). We attached an ear-tag with a unique identification number (Nasco, Atkinson, WI) on all captured Bobcats and stored the ear punch tissue in a sterile tube with 100% ethanol for subsequent DNA analysis. We fitted adult Bobcats weighing >9 kg with a global positioning system (GPS) collar (MiniTrack, Lotek Wireless Inc., Newmarket, ON, Canada) with a timer-released drop-off mechanism. The minimum weight requirement ensured that the GPS collar would be less than 4% of the body weight of the animal. We programmed the GPS collars to record a fix every 2 hours for 52 weeks. All trapping and animal handling procedures followed the guidelines of the American Society of Mammalogists (Sikes 2016) and the University of Rhode Island Institutional Animal Care and Use Committee protocol #AN1314-005.

Additional tissue samples used for genetic analyses were opportunistically collected from road-killed Bobcats located throughout Rhode Island during the study period (see Fig. S1 in Supplemental File 1, available online at http://www.eaglehill. us/NENAonline/suppl-files/n28-4-N1851-Mayer-s1, and for BioOne subscribers, at https://dx.doi.org/10.1656/N1851.s1).

DNA extraction and genotyping

We stored Bobcat tissue samples at -20 °C prior to DNA extraction. We extracted genomic DNA using either the DNeasy Blood and Tissue kit (Qiagen Sciences, MD) or the NucleoMag Tissue kit (Macherey-Nagel, Inc., PA) according to manufacturer's instructions. All DNA extractions and polymerase chain reactions (PCR) included negative controls to test for contamination in the reagents. We genotyped the DNA extracted from Bobcat samples using 12 microsatellite markers previously developed for Bobcat (BC1AT, BCD1T, BCE5T, and BCH6T; Faircloth et al. 2005) or the *Felis catus* L. (Domestic Cat; FCA023, FCA031, FCA045, FCA082, FCA149, FCA126, and FCA391 [Menotti-Raymond et al.

1999]; FCA740 [Menotti-Raymond et al. 2005]). We labeled the forward primers with a universal M13 sequence to allow for fluorescent dye labeling during PCR (Schuelke 2000). We labeled the reverse primers with a "pig-tail" by adding additional nucleotides to the sequence according to Brownstein et al. (1996). The PCRs contained 1 µl of genomic DNA; 2.5 units of Hotstart Master Mix (Qiagen Sciences); 0.25 μ M of forward primer; 0.5 μ M of the reverse primer; 0.5 μ M of the universal M13 primer labeled with either 6FAM (Integrated DNA Technologies, IA), NED (Applied Biosystems [ABI], CA), PET (ABI), or VIC (ABI); 0.14 µg/ µL of Bovine Serum Albumin (New England Biolabs, MA); 0.5 mM magnesium chloride; and PCR-grade water to make a final volume of 20 µl. We used a Mastercycler ep thermal cycler (Eppendorf North America, Enfield, CT) to amplify PCR products using 3 different thermocycler programs that differed by their annealing temperature. The PCR programs consisted of an initial denaturing step of 95 °C for 15 min; 30 cycles of 94 °C for 30 s, annealing temperature (56, 60, or 65 °C) for 45 s, and 72 °C for 45 s; 8 cycles of 94 °C for 30 s, 53 °C for 45 s, and 72 °C for 45 s; and a final extension of 72 °C for 10 min.

Amplified PCR products were cleaned using sera-mag beads (Rohland and Reich 2012) and prepared for fragment analysis at either Rhode Island Genomics and Sequencing Center (using an Applied Biosystems 3500xl Genetic Analyzer) or the Keck Biotechnology Resource Laboratory (using an Applied Biosystems 3730xL DNA Analyzer), according to the analyzers' instructions. We visualized and scored genotypes using the software Geneious Pro Version 9.1.8 (Biomatter, New Zealand). We repeated the genotyping of 6 samples (20% of our samples) that were run on the Applied Biosystems 3500xl Genetic Analyzer to estimate the allele error rate (Hoffman and Amos 2005). We also genotyped 4 samples using both an Applied Biosystems 3500xl Genetic Analyzer and an Applied Biosystems 3730xL DNA Analyzer to correct for any shifts in allele size scoring between the 2 platforms. We used the program Microchecker Version 2.2.3 (Van Oosterhout et al. 2004) to test for null alleles, genotyping errors, and departure from Hardy–Weinberg equilibrium.

Genetic diversity, effective population size, and population structure

We estimated unbiased expected heterozygosity and mean pairwise relatedness (Lynch and Ritland 1999) using GenAlEx v.6.5 (Peakall and Smouse 2012). We estimated contemporary effective population size (N_e) with a minor allele frequency of 0.05 using the co-ancestory method of Nomura (2008) implemented in NeEstimator V2.1 (Do et al. 2014). We used an exploratory data analysis method to estimate population structure by conducting a principal coordinate analysis (PCoA) in GenAlEx v.6.5. We also used a model-based clustering approach to estimate population structure using the Bayesian program Structure (Pritchard et al. 2000). We conducted 10 independent runs with a burn-in period of 100,000 and 200,000 subsequent MCMC iterations. We implemented the admixture module with correlated allele frequencies and tested a priori cluster values (K) from 1 to 4. We used the program Structure Harvester (Earl and VonHoldt 2012) to select the optimal K.

GPS collar accuracy

Prior to deploying the GPS collars on any Bobcats, we evaluated location accuracy by placing collars in 4 different land-cover types with associated canopyclosure classes (open: 0-25%, edge: 26-50%, forest: 51-75%, dense understory forest: 76-100%). We placed the collar around a bottle filled with saline solution following Forin-Wiart et al. (2015) and placed the bottle on a stand ~44 cm high to simulate the average height of a Bobcat. During testing, the collars were programmed to collect a GPS location every 20 minutes for at least 24 hours. We calculated the distance from each recorded point to the actual location of the collar and averaged across all habitat types to get an estimate of overall GPS accuracy.

Home range

We estimated home-range and core area using the 95% and 50% utilization distribution (UD) from a fitted autocorrelated kernel density estimator (AKDE; Fleming and Calabrese 2017, Fleming et al. 2015). We removed the first 2 weeks of GPS locations from each collared individual to account for any behavioral abnormalities during the recovery time post-capture (Northrup et al. 2014) and subsetted data in order to calculate the annual, summer (15 May–14 October), and winter (15 October-14 May) home ranges for each individual. The summer and winter cut-off dates corresponded with approximate broad-leaved deciduous leaf-on and leaf-off dates in Rhode Island. We evaluated the subsetted data sets for outliers using the outlier() function in the R package 'ctmm' (Calabrese et al. 2016) in RStudio using R version 3.3.3 (R Development Core Team 2017, RStudio Team 2016). We identified and removed the top 5% of GPS fixes that were furthest from the median location for each individual and then fit the remaining data points separately using the R package 'ctmm' (Calabrese et al. 2016). We restricted the UD extent to the mainland of Rhode Island by creating barriers along the coastline to control for overestimation of the home range.

Resource selection

We used a third-order resource-selection analysis (Johnson 1980) within a usedavailability framework (Manly et al. 2002) for evaluating the resource selection of individual Bobcats within their home range. We used the R-package 'amt' to prepare location and covariate data for analysis (Signer et al. 2019) and the random_points() function to derive the availability sample by systematically spacing "available" points at a ratio of 100 points for each used location. We then removed any "available" points that fell in areas that were non-habitat and thus unavailable to a Bobcat (e.g., inland water bodies and coastal salt ponds); we conducted a sensitivity analysis in regard to the size of the available sample to ensure model parameters were estimated appropriately (Northrup et al. 2013).

Previous studies on Bobcat resource selection identified the importance of certain land-class types, including wetlands, shrublands, and forests (Broman et al. 2014, Clare et al. 2015), and topographic covariates, such as elevation and slope (Broman et al. 2014, McCord 1974), in describing selection. As such, we hypothesized that these land-class types would be important in identifying resource

selection in our study area. Furthermore, we hypothesized that anthropogenic features would influence Bobcat selection because road density is negatively associated with Bobcat occurrence in other areas (Broman et al. 2014) and included them in our analysis to characterize Bobcat response to high human population density and development. Fine scale geographic information system (GIS) datasets for land class and young forests were only available for Rhode Island with no directly comparable datasets available for nearby states, so all resource-selection analyses were restricted to within the borders of Rhode Island. We identified 9 resource covariates to include in the analysis based on prior studies and available GIS datasets (Table 1). We simplified land-class types from 38 descriptions in the original ecological communities dataset to 7 broad land class categories (agriculture, forest, grassland, forested wetland, other wetland, shrubland, and developed). We evaluated the correlation among resource covariates by generating a matrix of Pearson correlation coefficients and generated variance inflation factors for each covariate from a global model of all covariates. There was moderate correlation between "distance to roads" and 2 other variables (road density and distance to edge), so we

Variable	Description and justification	Source
Land cover class	Agriculture, forest, shrubland, grassland, wetland, forested wetland, or developed. Bobcats select certain habitat types over others (Broman et al. 2014)	RI Ecological Communities Classification
Distance to wetlands (m)	Wetland habitats and wetland edges provide high diversity of prey species (Stephens and Anderson 2014) and have been found to be associated with increased Bobcat densities (Clare et al. 2015)	RI Ecological Communities Classification
Distance to roads (m)	Roads have a negative impact on survival (road mortality) and may be avoided (Broman et al. 2014)	RIDOT Roads
Road density	Kilometers of road per km ² . Roads have a negative impact on survival (road mortality) and may be avoided (Broman et al. 2014)	RIDOT Roads
Distance to young forest (m)	Young forests (scrub/shrub) are preferred habitat of cottontails (prey source) and provide cover (Fuller and DeStefano 2003, Litvaitis 2001)	Young Forest in RI 2018 v2 (available at www. arcgis.com)
Distance to edge habitat (m)	20-m buffer zone around edge of where areas that offer cover meet open areas. Edge habitats act as travel corridors and typically have high prey densities (Abouelezz et al. 2018, Harper et al. 2005)	RI Forest Habitat (2010) classified as open or cover
Aspect (degrees)	Bobcats have been shown to avoid NW aspects in northern New England (Broman et al. 2014)	2011 statewide (LiDAR)
Slope (degrees)	Bobcats are associated with rugged slopes and ledges in New England (McCord 1974)	2011 statewide (LiDAR)
Elevation (m)	Bobcats are associated with lower elevations in northern New England (Broman et al. 2014)	2011 statewide (LiDAR)

Table 1. Description, justification, and data source of variables used to investigate Bobcat resource selection within a home range in Rhode Island. All data, unless noted, can be accessed through RIGIS.org.

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removed this covariate from further analysis. We categorized the remaining covariates as either habitat type (land class), distance to habitat (distance to wetland and distance to young forest), anthropogenic feature (road density), fragmentation feature (distance to edge), or topographic (aspect, slope, and elevation). We generated 26 a priori models with combinations of covariates (see Table S1 in Supplemental File 1). Eight models had a single variable and the remaining 18 models contained land class plus variables from at least 1 additional category; the most complex model considered 14 variables.

We fit RSFs by approximating a spatial point process model using logistic regression (Hooten et al. 2017), fitting models using the glm() function in R for each individual Bobcat separately. We focused on individual-level analyses because selection is expected to vary by individual and is thus the primary unit of interest in RSF analyses (Montgomery et al. 2018). We used Akaike's information criterion (AIC) (Akaike 1973) to assess model parsimony and identified the most supported models using AIC weights (Burnham and Anderson 2002). We used the AIC weights and coefficients from each model to derive model-averaged predictions of resource selection across the home range, seasonally for each individual. We produced RSF predictions for each model using the estimated coefficients and relevant covariates after removing the intercept and exponentiating the values (Hooten et al. 2017). We evaluated goodness-of-fit using k-fold cross-validation and calculating spearman rank correlation and a *P*-value ($\alpha = 0.05$) per fold, following Boyce et al. (2002).

Results

We had a total of 2232 trap nights across 4 field seasons and captured 8 Bobcats (6 male, 2 female). Two additional Bobcats (1 male, 1 female) were captured by recreational trappers as by-catch outside of the trapping study area, but were processed and included in this study. The average total length (head to tail) for captured females and males was 73.5 cm (SE = 3.9) and 83.7 cm (SE = 2.9), respectively, and the average weight for females and males was 6.4 kg (SE = 0.2) and 9.2 kg (SE = 1.0), respectively. All females and 1 male were under the minimum weight requirement for the GPS collars and thus were only measured, fitted with an ear-tag, and released. Four males were fitted with GPS collars; of these, 1 collar malfunctioned and no location data was collected for that individual. Fix rate success was high for the 3 functioning GPS collars (mean = 79.9%, SE = 5.5), and a sufficient number of locations were collected from these individuals to be able to complete a seasonal home-range and resource-selection analysis (467–1306 locations per season). One individual's collar was only active for the winter season (108 days), while the others were active during both the winter and summer seasons (261–293 days).

Genetic diversity, effective population size, and population structure

We obtained genetic samples from a total of 30 individual Bobcats: 10 trapped, 18 road-mortality, and 2 confiscated from illegal harvesting (see Table S2 and Fig. S1 in Supplemental File 1). Our genotype error rate was 3.3% (2 mismatches out of

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61 genotype comparisons), and our allele error rate was 1.6% (2 allele mismatches out of 122 allele comparisons). The 4 samples that were analyzed on each Applied Biosystems platform had matching alleles after a consistent shift in size by either 1 or 2 base pairs, but 5 genotypes out of the 96 comparisons failed to amplify. We found no evidence of null alleles, genotyping errors, or departure from Hardy–Weinberg equilibrium.

The mean unbiased expected heterozygosity was 0.763 (SE = 0.020), while the values for each marker varied from 0.625 to 0.852 (Table 2). The overall mean relatedness was -0.017 \pm 0.003 SE with a min–max of -0.155–0.303 (see Table S3 in Supplemental File 1). When looking at just the collared Bobcats from this study, the pairwise relatedness values showed they were not close relatives to each other, with a maximum relatedness value of 0.049. Bobcat M01 was most closely related to RI-BC-002 (0.170), M03 was most closely related to RI-BC-09 (0.120), and M04 was most closely related to RI-BC-001 (0.130). The N_e was estimated at 82 (95% confidence interval: 44–329).

Both the PCoA and structure analyses indicated no population structure. The PCoA showed a scattering of the individuals with no geographic pattern and 10.5% and 9.8% variation explained by the first 2 principal coordinates, respectively (see Fig. S2 in Supplemental File 1). Structure Harvester determined that delta K was greatest for a cluster size (K) of 3; however, every individual had an equal proportion of membership in the 3 clusters (Fig. 1). Thus, the mean natural log probability of the data was a more appropriate measure to identify K, which peaked at K = 1.

GPS collar accuracy

We found the average collar accuracy across all tested cover types was 10.2 m (SE = 0.5) Within each cover type, the average collar accuracy was 7.4 m (SE = 0.7) in the "open" cover class, 11.9 m (SE = 1.8) in the "edge" cover class, 12.8 m

Table 2. Genetic diversity summary for 12 microsatellite markers estimated using GenAlEx v.6.5 (Peakall and Smouse 2012). Sample size (*n*), number of different alleles (N_a), number of effective alleles (N_e), Shannon's information index (I), observed heterozygosity (H_o), expected heterozygosity (H_e), unbiased expected heterozygosity (uH_e), and Fixation index (F)

Locus	n	$N_{\rm a}$	$N_{\rm e}$	Ι	H _o	H _e	uH _e	F
FCA031	30	6.000	4.580	1.643	0.800	0.782	0.795	-0.023
FCA149	30	8.000	4.380	1.742	0.733	0.772	0.785	0.050
BCIAT	30	5.000	3.416	1.399	0.700	0.707	0.719	0.010
BCH6T	29	7.000	6.139	1.870	0.793	0.837	0.852	0.053
BCD1T	30	4.000	3.056	1.158	0.700	0.673	0.684	-0.040
FCA740	30	7.000	5.294	1.752	0.833	0.811	0.825	-0.027
FCA082	30	6.000	4.545	1.612	0.967	0.780	0.793	-0.239
FCA023	30	6.000	4.523	1.593	0.867	0.779	0.792	-0.113
FCA045	30	5.000	2.594	1.188	0.733	0.614	0.625	-0.193
BCE5T	30	5.000	2.922	1.241	0.633	0.658	0.669	0.037
FCA126	29	6.000	4.933	1.688	0.966	0.797	0.811	-0.211
FCA391	30	6.000	4.865	1.663	0.800	0.794	0.808	-0.007

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(SE = 1.4) in the "forest" cover class, and 9.9 m (SE = 0.6) in the "dense understory forest" cover. For subsequent home-range analyses, we incorporated collar error within the AKDE by setting the user equivalent range error to 10 m and did not filter any locations out of the collected telemetry datasets.

Home range

We found the average winter home-range and core-area sizes were 219.3 km² (SE = 73) and 51.7 km² (SE = 3.2), respectively (n = 3; Table 3). The average



Figure 1. Population genetic structure of 30 Bobcat individuals genotyped at 12 microsatellite markers using the program Structure (Pritchard et al. 2000). We conducted 10 independent runs with a burn-in period of 100,000 and 200,000 subsequent MCMC iterations. We implemented the admixture module with correlated allele frequencies and tested a priori cluster values from 1 to 4. The x-axis numbers represent each individual, which corresponds sequentially to our Bobcat sample numbers. The y-axis represents the proportion membership in 1 of 3 clusters.

Table 3. Home range (95% UD) and core-use area (50% UD) for individual Bobcats in Rhode Island by season (Winter: 16 October–15 May, Summer: 16 May–15 October). Home-range estimates were calculated using a fitted autocorrelated kernel density estimator. The top 5% of outliers were identified and removed prior to calculating home range for each individual and each season. Note that for M03, not enough fixes were obtained during the first winter season to complete a seasonal analysis and so are not included in the count under winter home range, but those fixes were retained in the annual analysis.

	Annual	home ran	ge (km ²)	Winter	home rang	(km^2)	Summer l	nome ran	ge (km ²)
ID	<i>n</i> fixes	95%	50%	<i>n</i> fixes	95%	50%	<i>n</i> fixes	95%	50%
M01	-	-	-	858	234.83	69.68	-	-	-
M03	1943	150.28	34.40	467	85.75	26.06	1193	92.30	18.83
M04	2540	285.84	51.10	1306	337.19	59.47	1234	56.71	17.39

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summer home-range and core-area sizes were 74.5 km² (SE = 17.8) and 18.1 km² (SE = 0.7), respectively (n = 2). The 2 individuals that had analyses for both seasons displayed a shift in the location of the core-use areas between seasons (Fig. 2).

Resource selection

Given the few individual Bobcats that were able to be sampled, it is difficult to generalize our findings of resource selection to the entire population of Bobcats in Rhode Island. Nonetheless, we can learn about resource selection from these individuals by comparing and contrasting findings among these individuals and other results from New England. Because there are relatively few RSF studies in New England and currently none in southern New England, these results are an important contribution to building our understanding of the landscape attributes that influence the behavioral decision-making and thus fitness effects on Bobcat. Here, we found 5 of the same resource covariates were included in the most supported models for each seasonal RSF model: land class, distance to young forests, distance to wetlands, road density, distance to edge, and slope (Table 4). We also found that aspect was included in 2 of the individual RSF models (M04 winter and M03 summer) and elevation was included for all but 1 of the models (M03 winter) (Table 4). There was no indication of a lack of model fit for any RSF model (see



Figure 2. Individual annual and seasonal home ranges for Bobcats tracked in southern Rhode Island from winter 2014 to winter 2018. Home ranges were estimated using a fitted autocorrelated kernel density estimator.

lable 4. Model coefficients (b) and ar season (Winter: 16 October–15 May, from 2015 to 2018.	ssociated standard error Summer: 16 May–15 O	(SE) tor each covariate ctober). Models were fi	the most parsimonic tusing data collected f	ous RSF model for each rom 3 GPS-collared Bc	i individual Bobcat by bcats in Rhode Island
Covariate	M01 - Winter	M03 - Winter	M04 - Winter	M03 - Summer	M04 - Summer
Intercept (land class – grassland)	-16.28 (0.28)	-13.63 (0.25)	-13.33 (0.12)	-13.07 (0.12)	-12.79 (0.20)
Land class-shrubland	1.23 (0.27)	$0.47~(0.27)^{*}$	$0.15 (0.14)^{*}$	0.76(0.13)	1.08 (0.22)
Land class- forest	0.92 (0.27)	$0.38~(0.26)^{*}$	-0.35 (0.13)	$0.03 \ (0.13)^{*}$	-0.44(0.20)
Land class- agriculture	$0.63\;(0.38)^{*}$	$0.55\ (0.38)^{*}$	-0.20(0.19)	-1.17 (0.30)	$-0.29(0.48)^{*}$
Land class- developed	$0.21 \ (0.27)^{*}$	-0.60 (0.28)	-0.88 (0.14)	-1.57 (0.16)	-1.07 (0.28)
Land class- wetland	$-0.08(0.38)^{*}$	$-0.36 (0.56)^{*}$	0.44 (0.22)	-0.87 (0.38)	
Land class - forested wetland	1.27 (0.27)	1.11 (0.27)	0.31(0.14)	0.71 (0.15)	$0.11 (0.24)^{*}$
Distance to young forest	-1.56(0.11)	-0.22 (0.06)	-1.03 (0.07)	-0.37(0.04)	-0.77 (0.11)
Distance to wetland	-0.26 (0.05)	-0.62 (.10)	-0.19 (0.04)	0.17(0.03)	$-0.12(0.09)^{*}$
Road density	0.42(0.03)	$0.04~(0.05)^{*}$	0.49(0.04)	0.16(0.03)	$0.06\ (0.13)^{*}$
Distance to edge	-0.78 (0.09)	0.16(0.07)	-0.39 (0.06)	0.30(0.04)	$-0.12(0.08)^{*}$
Aspect			-0.08(0.03)	-0.10(0.03)	
Slope	0.20(0.03)	$0.09\ (0.05)^{*}$	0.13(0.03)	-0.09 (0.03)	$0.003 \ (0.06)^{*}$
Elevation	-1.45 (0.09)		-0.64(0.05)	$0.06~(0.04)^{*}$	-0.25 (0.07)
$^*P > 0.05$					

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Downloaded From: https://bioone.org/journals/Northeastern-Naturalist on 27 Dec 2023 Terms of Use: https://bioone.org/terms-of-use Access provided by Colorado State University Table S4 in Supplemental File 1). We used all models for each individual with AIC weights >0.001 (see Table S5 in Supplemental File 1) to create the model-averaged RSF maps for each individual seasonally (Fig. 3).



Figure 3. Individual model-averaged resource selection function (RSF) maps for Bobcats in Rhode Island. (A) Locations of seasonal home ranges for each individual Bobcat tracked in southern Rhode Island. Remaining panels show RSF map for (B) M01 in winter, (C) M03 in summer, (D) M03 in winter, (E) M04 in summer, and (F) M04 in winter.

Shrublands and forested wetlands were the only 2 land-class resource covariates that were selected for more than their availability by each individual in each season. Other wetland types that included emergent marshes, peatlands, and salt marshes were mostly avoided or not used by most individuals, as was the developed land-class type. Distance to young forests also was an important variable, with all individuals across seasons consistently selecting areas closer to this habitat type. Selection/avoidance for resources characterizing "fragmentation" were inconsistent among individuals and seasons. Selection for areas close to edge habitats also was inconsistent among individuals; however, each individual had higher relative probability of selection for areas with higher road densities. Two individuals showed higher relative probability of selection with lower degrees of aspect (northerly aspects). All individuals had a higher relative probability of selection in areas with steeper slopes at lower elevations for at least 1 season. There was variation in resource selection among individuals when comparing selection coefficients during the winter season in particular. Despite overlap of the core-use area of each individual (Fig 1.), there were inconsistencies in selection or avoidance of the forest, agriculture, development, wetland, and distance-to-edge variables (Table 4).

Discussion

Bobcats are widely distributed, opportunistic carnivores that make use of diverse land-cover types throughout their range. With no prior studies on Bobcats in southern New England, it is difficult for resource managers to employ evidencebased management practices with regard to a species that is so highly adaptable. Therefore, we aimed to understand the spatial ecology of Bobcats in Rhode Island by characterizing their population genetic structure, as well as estimating ranging patterns and resource selection. Not surprisingly, Bobcat in Rhode Island represented a single contiguous genetic population. Previous research on Bobcats in the northeastern US have detected population structure, but their study was at a much larger scale (Carroll et al. 2019). We detected the movement of Bobcat over a span greater than half the length of Rhode Island, which shows their ability to easily be connected across the entire state. Surprisingly, Bobcats in Rhode Island have retained a relatively high level of genetic diversity and effective population size, and our collared Bobcats were not close relatives to each other. Our estimated unbiased expected heterozygosity was higher than the genetic diversity estimates from New Hampshire and Vermont populations (Carroll et al. 2019), but the markers used in each estimate are not the same, so caution is needed for these comparisons. Our $N_{\rm e}$ estimate is the first relative population-size estimate for Rhode Island Bobcat, and the value is higher than $N_{\rm e}$ estimates for Bobcat in eastern New Hampshire and Vermont lowlands (Carroll et al. 2019). However, caution also is needed for these comparisons because each study used a different number of markers and methods to estimate $N_{\rm e}$ and we likely did not sample from the full extent of the population in our area. The confidence interval on our N_e estimate also is fairly large.

We predicted that the home-range size and resource selection of Bobcats in our study would be larger than those in other regions, due to the unique features of

southern New England and the resulting differences in resource availability. The average male Bobcat home-range size in our study (218.06 km²) was larger than what was reported in studies from other regions of North America, such as Wisconsin (65.9 km², n = 6; Lovallo and Anderson 1996), Kansas (20.0 km², n = 1; Kamler and Gipson 2000), and California (6.78 km², n = 12; Poessel et al. 2014), as well as in more northern portions of New England such as Maine (71.1 km² [n = 4] and 112.2 km² [n = 6]; Litvaitis et al. 1986), Vermont (70.9 km², n = 10; Donovan et al. 2011), and New Hampshire (93.5 km², n = 10 [Broman et al. 2014]; 81.6 km², n = 13 [Reed et al. 2017]).

There was some evidence in the movement data to suggest that 1 individual (M01) did not have a resident home range but was perhaps in a transient phase. Typical transient behavior in Bobcats includes erratic patterns of space use that is not limited to a well-defined home range (Benson et al. 2004, Litvaitis et al. 1987). While this particular individual had a defined core-use area, there were multiple instances of long-distance exploratory movements where the individual strayed far from the core area for weeks at a time. While some of these data points were removed as outliers prior to home-range estimation, this transient behavior of 1 individual could partially explain the higher average home-range estimates; however, the other non-transient individuals in the study had similar home-range sizes during the same season. The 2 individuals that were followed for 2 seasons both exhibited shifts in their home range on a seasonal basis. This was most evident in individual M04, whose core-use area was divided between coastal RI and interior western Rhode Island/eastern Connecticut in winter but shifted to only the interior region in the summer. Both individuals had smaller home ranges and core-use areas during the summer than in the winter. In female Bobcats, a decrease in the summer home-range size has been shown to be related to behavioral changes due to reproduction (Kamler and Gipson 2000), but an increase in winter home-range size in both males and females may indicate a shortage of resources during the winter requiring more movement to fulfill their biological needs (Litvaitis et al. 1987).

Using resource-selection functions to characterize an animal's resource requirements provide resource managers with information specific to a species' home range, allowing for informed decision-making in terms of land conservation and habitat management on a local scale. Focusing on this local scale is important due to variations in resource availability across larger areas, as well as variation in habitat preferences within a particular population (Smith and Schaefer 2002). We specifically found individual-level Bobcat resource selection was similar to results from previous studies, where wetland and shrubland habitats and low-elevation areas had high relative-selection probabilities; however, contrary to the findings of other studies, we found areas with high road density were not avoided (Broman et al. 2014, Donovan et al. 2011, Reed et al. 2017). In New Hampshire, Reed et al. (2017) found that areas with high road density were largely avoided by Bobcats, while each individual in our study had positive selection coefficients for areas with higher road densities. Studies in other landscapes have noted similar avoidance responses

to roads (e.g., Riley 2006); however, individual variation in the amount of use of areas near roads is noted. When examining the habitat types within the land-class variable, developed areas were generally avoided. This result suggests that Bobcats are not necessarily selecting for areas with a high anthropogenic footprint, but are more likely utilizing the habitats and resources that are typically associated with road edges. There were strong associations with edge-type habitats-the buffer area between forest cover and open areas—as well as areas closer to wetlands and young forest habitat. These edge-type habitats are often found near roads, which may have had an influence on the selection for areas with higher road density. Additionally, the area where all of the collared animals were captured, southern Rhode Island, is a densely populated area. To more fully understand Bobcat selection of habitat near roads, a finer-scale movement-based analysis of how Bobcats travel across roads (e.g., GPS fix every 10 minutes), along with a comparison of individuals' selection of resources in areas of both higher and lower road density than found in our study area is warranted. We further observed minimal seasonal differences in resource selection, with only 1 individual exhibiting a switch from selection to avoidance of agricultural areas between winter and summer, and a change in selecting areas closer to wetlands during the summer compared to winter. This finding is in contrast to the shift in space use between the seasons, which suggests that as the availability of resources changes across seasons, Bobcats may have to actively seek out new areas to meet their resource requirements.

There were significant limitations in this study due to small sample size. Despite a large trapping effort, there were few captures, and the majority of individuals captured were under the minimum weight requirements for the GPS collars. This low trap success speaks to the likely low population densities of Bobcats in the study area. However, the 3 individuals that were successfully collared and tracked provided sufficient data for robust individual home-range and resource-selection analyses. We acknowledge that our small sample size, comprised of only males, limits our ability to apply our findings across the entire population of Bobcats in the state, and we are cautious about making broad management guidelines based on this small sample size. However, these data and analyses provide wildlife managers information on a species that has not been studied in the region previously and provides baseline ecological information on this potentially increasing population. On a broader scale, the data presented provides insight on how Bobcats can successfully use an altered landscape as resource availability is shifting due to higher densities of humans and roads.

Our study highlights the importance of examining the ecology of a widely distributed and adaptable species on a local scale (i.e., Rhode Island). While we found some similarities between the spatial ecology of Bobcats in Rhode Island and other regions, we also found notable differences. We encourage local conservation and land management groups interested in supporting Bobcat populations to consider not only conserving important habitats, such as wetlands and young forests, but the habitats surrounding these areas to create protected edge habitats. Furthermore, given that Bobcats likely represent a single population within the

state of Rhode Island, any future population management should be considered at this scale or larger. Lastly, considering the large-scale potential movements of Bobcats, their low densities, and the relative size of southern New England states, multi-state collaboration may need to be considered in future landscape-management planning.

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