GENETIC STRUCTURE AND DIVERSITY IN BOBCAT (*LYNX RUFUS*) POPULATIONS IN OKLAHOMA: PRELIMINARY RESULTS

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Abstract

The bobcat (*Lynx rufus*) has a wide distribution across North America, and is found in a variety of habitats. In many areas, including Oklahoma, the species is harvested for its pelt, with seasonal hunts on a yearly basis. There is concern that the removal of too many individuals could lead to an increase in inbreeding, as a result of small population sizes, with the possibility of alterations to the populations physical and genetic health. Specimens collected from across the state during different annual hunting seasons will be compared to each other to determine any possible notable variations within the genetic diversity of bobcats in Oklahoma as a result of the harvest. To date, DNA has been extracted from 287 samples collected during a single harvest year, and preliminary data has been generated for 5 of 19 microsatellite loci for 10 of these samples.

Bobcat Population Structure

Bobcats are solitary and are only found together when mating or rearing young (Cochrane et al., 2006). Adult male bobcats will move away from their mother's home range to establish a territory that they will defend (Sculley et al., 2018). Females are philopatric and tend to remain in a single territory, rarely changing their home range during their lifetime. However, they are known to move into a new territory on occasion, such as when another adult female dies and leaves a territory empty (Lovallo and Fowles, 2018). Mating between dispersing bobcats also allows for the maintenance of genetic variation within the species and can limit the effects of inbreeding (Schwartz et al., 2003). However, due to the 1:1 sex ratio seen in most populations and male-biased dispersal, there is concern that the removal of male bobcats might have a negative effect on the genetic health of some populations (Johnson et al., 2006).

Annual Harvest and Genetic Diversity

Bobcat pelts carry a high monetary value, and are sought after by hunters and trappers for the creation of clothing and decorative items, such as hunting trophies (Lavoie et al., 2009). In Oklahoma, the season goes from 1 December to 28 or 29 February, with a limit of 20 bobcats per season per trapper. The season usually leads to the removal of between 9-10 thousand individuals from the state's total population (Chrisman et al., 2019), with a total of 10,506 bobcats being trapped in the 2018-2019 season alone. There is concern that the removal of large numbers of individuals from a population could severely impact the health of species, due to an increase in inbreeding and the possibility of an increase in detrimental mutations (Millions and Swanson, 2005), and resulting in cascading effects within environments, such as the expansion of other mammals and reduction of plant life (Petraborg and Gunvalson, 1962). The objective of this research is to determine the genetic health and population structure of bobcats through the use of trapper harvested tissue samples.

Tissue Collection

A total of 287 tissue samples were collected from bobcat specimens from 41 of 77 counties during the 2018 to 2019 season. Samples were obtained from all geographic regions of Oklahoma, including the panhandle. Tongues were removed from each bobcat by the Oklahoma Department of Wildlife Conservation and deposited at Oklahoma State University for parasite analyses. Tissue subsamples were taken from the tongues and DNA was extracted with the use of DNeasy Tissue Extraction Kits. Samples were quantified using a Nanodrop-2000.

PCR and Genotyping

- Tissue samples are being amplified with 19 microsatellite loci. To date, 15 loci have been optimized (Table 1).
- Samples are being genotyped on a 3500 Genetic Analyzer. To date, preliminary data has been generated for 5 loci (Table 2).
- Preliminary diversity statistics were generated using Cervus 3.0.7 (Marshall et al., 1998; Slate et al., 2000; Kalinowski et al., 2007, 2010; Table 3).

Table 1.—Microsatellite primers to be used in this study to uniquely identify individual bobcats (*Lynx rufus*). The primer name, citation source, expected size range, and optimal annealing temperature are provided. BC = primers developed using bobcats, Fca = primers developed using domestic cats (*Felis catus*), and Lc = primers developed using Canada lynx (*L. canadensis*). All primers have been shown to work in bobcats.

Primer Name	Source	Allele Size Range	Annealing Temp. (°C)
BC1AT	Faircloth et al. 2005	318	50-60
BCE5T	Faircloth et al. 2005	261	50
BCG8T	Faircloth et al. 2005	265	48-54
Fca008	Menotti-Raymond and O'Brien 1995	122-146	56
Fca031	Menotti-Raymond et al. 1999	221-241	52
Fca035	Menotti-Raymond et al. 1999	136-150	52
Fca077	Menotti-Raymond et al. 1999	143-155	40-62
Fca082	Menotti-Raymond et al. 1999	238-246	54
Fca090	Menotti-Raymond and O'Brien 1995	93-120	58-60
Fca096	Menotti-Raymond et al. 1999	184-224	53
Fca126	Menotti-Raymond et al. 1999	139-145	58-60
Fca132	Menotti-Raymond et al. 1997	137-153	54-60
Fca391	Menotti-Raymond et al. 1999	237-273	56-58
Fca559	Menotti-Raymond et al. 1999	141-195	
Fca740	Menotti-Raymond et al. 2005	199-223	58-60
Fca742	Menotti-Raymond et al. 2005	123-175	
Lc109	Carmichael et al. 2000	172-182	
Lc110	Carmichael et al. 2000	91-103	50-56
Lc118	Carmichael et al. 2000	133-145	

Table 2.—Allele calls for 10 bobcats (BC) for 5 microsatellite loci. "a" and "b" indicate the 2 alleles for each locus. N/A indicates that no data currently is available.

Sample	LC110a	LC110b	BC1ATa	BC1ATb	FCA90a	FCA90b	BCE5Ta	BCE5Tb	BCG8Ta	BCG8Tb
BC022	74	82	306	310	107	109	256	272	280	286
BC023	74	74	304	306	N\A	N\A	264	264	282	295
BC024	78	82	314	322	N\A	N\A	256	276	295	295
BC025	74	82	306	318	N\A	N\A	264	268	282	282
BC027	78	82	310	318	N\A	N\A	258	268	282	295
BC028	78	82	310	314	N\A	N\A	264	276	291	297
BC029	78	78	310	314	N\A	N\A	268	276	291	297
BC030	78	78	310	318	N\A	N\A	N\A	N\A	291	295
BC031	74	82	304	306	109	109	268	272	295	299
BC032	N\A	N\A	302	307	N\A	N\A	264	264	N\A	N\A

Table 3.—Preliminary diversity indices estimated using Cervus. Locus, number of individuals genotyped for each locus (N), number of alleles per locus (k), observed heterozygosity (H_O), expected heterozygosity (H_E), and polymorphic information content (PIC) are provided.

Locus	N	k	H_O	H_E	PIC
LC110	9	3	0.667	0.669	0.586
BC1AT	10	8	1	0.879	0.814
FCA90	2	2	0.500	0.500	0.305
BCE5T	9	6	0.778	0.830	0.753
BCG8T	9	7	0.778	0.837	0.762

Future Work

We are in the process of optimizing the remaining 4 loci. Additionally, we are optimizing a Y chromosome STR to allow us to determine the sex of each cat because that data was not provided when samples were obtained. Currently, 20 additional samples have been amplified for all optimized loci and we are awaiting data from the genetic analyzer runs. We are awaiting samples from the 2019-2020 trapping season to allow comparisons between at least 2 seasons. This comparison will allow us to determine the impact of these two harvest seasons on genetic diversity in the population. Additionally, we will be examining genetic structure in Oklahoma populations to help inform management decisions regarding this species.

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