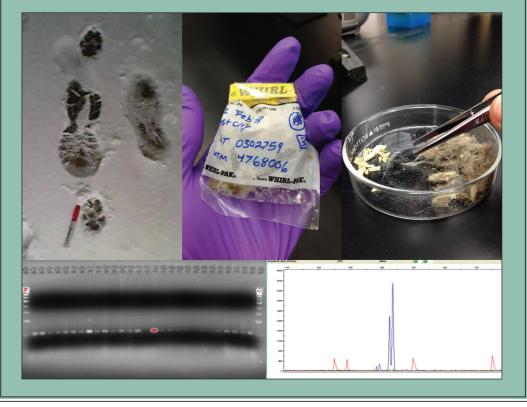
# Urban Naturalist

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## Diet, Habitat Use, and Size of an Urban Population of Coyotes Obtained Noninvasively

Tyler Garwood, Ross Ziobro, Kathleen Keene, Angelina Waller, and Jonathan N. Pauli



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**Cover Images:** Non-invasive approaches—scat collection, dietary analysis, and multilocus genotyping—to quantify Coyote a population in an urban landscape. Images © Tyler Garwood, Ross Ziobro, Kathleen Keene, Angelina Waller, and Jonathan N. Pauli.

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#### Diet, Habitat Use, and Size of an Urban Population of Coyotes Obtained Noninvasively

Tyler Garwood<sup>1</sup>, Ross Ziobro<sup>1</sup>, Kathleen Keene<sup>1</sup>, Angelina Waller<sup>1</sup>, and Jonathan N. Pauli<sup>1,\*</sup>

**Abstract** - Anthropogenically altered landscapes are novel ecosystems that some species successfully exploit. *Canis latrans* (Coyote) can attain high densities in urban ecosystems and is of public interest because of its perceived conflict with humans. Using a combination of noninvasive scat sampling, multilocus genotyping, and closed-population modeling, we determined the population size of Coyotes within a natural area in Madison, WI. We estimated a population size of 10–19 individuals at the site (~0.02 Coyotes/ha), and although Coyotes strongly selected for horticultural cover types (mainly non-native plants), their diets were principally comprised of native small-mammal prey. These findings support the notion that Coyotes can reach relatively high densities in urban areas, and may exploit anthropogenically altered habitats while still relying primarily on native prey items.

#### Introduction

Numerous generalist species have benefited as the North American landscape has become increasingly developed (Thurber and Peterson 1991, Waller and Alverson 1997). One of these species, *Canis latrans* Say (Coyote), has spread eastward and now thrives in heavily urbanized areas, a landscape previously assumed inhospitable to a predator of its size (Rashleigh et al. 2008). Coyotes persist and reach relatively high densities in urban habitats by denning in or near forest preserves, city parks, and wooded residential areas (Gese et al. 2012). These animals are able to exploit diverse food types available in urban habitats (Kozlowski et al. 2008), including human-derived items (Todd and Keith 1983).

Urban Coyote populations are also of particular public interest because of potential Coyote-human conflicts. Indeed, in the Chicago metropolitan area, which features a large and growing Coyote population, Coyotes rank as the species of wildlife perceived as the greatest threat to human safety (White and Gehrt 2009). Certain Coyote behaviors, including moving through areas of high human density at night when they are less likely to encounter people and selecting for natural areas such as forest preserves within the urban landscape, minimize conflict (Gese et al. 2012, Grubbs and Krausman 2009). However, other behaviors, including an inclination for use of human-made trails during heavy snow cover (Whiteman and Buskirk 2013) and adaptable foraging strategies, could make Coyotes more likely to come into contact with humans and pets. A recent review suggested that the number of Coyote attacks on pets has risen in recent years (White and Gehrt 2009).

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In southern Wisconsin, Coyotes were historically rare until at least the 1920s (Schorger 1973). However, increases in Coyote abundance have been noted in metropolitan areas across the Midwest (Gehrt 2004), and anecdotal observations suggest that this is also true for urban areas in southern Wisconsin. However, little is known regarding the size, habitat preference, or dietary composition of current Coyote populations in Madison, WI, and other regional mid-sized metropolitan areas. Our objectives were to quantify the population size and diet of urban Coyotes and determine their habitat use within an urban system. Based on previous studies (Gehrt 2004), we predicted that the Coyote density in natural areas of Madison, WI, could be high and that Coyotes would mostly select for well-structured, natural cover types (Gese et al. 2012). We also expected that although concerns regarding conflicts with pets and humans are high, Coyotes' primary prey items would be native small mammals (Newsome et al. 2015).

#### **Field-Site Description**

We conducted our study in the University of Wisconsin-Madison Arboretum, a natural area surrounded by dense housing developments, businesses, and industry within the city limits of Madison, WI. We chose to sample 12.5 km of trails that included 6 distinct cover types within the arboretum: coniferous forest (*Pinus resinosa* Aiton [Red Pine] and *P. strobus* L. [White Pine]), deciduous forest (closed canopy *Quercus* spp. [Oak]), horticulture (ornamental trees and shrubs), oak savanna, wetlands, and tallgrass prairie (Fig. 1; M. Wegener, University of Wisconsin-Madison Arboretum, Madison, WI, unpubl. data).

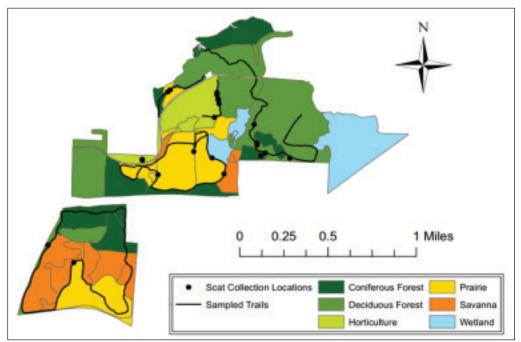


Figure 1. Location of sampled trails, scat-sample locations, and habitats within the University of Wisconsin-Madison Arboretum, winter 2013–2014.

#### Methods

We collected scat samples every 4 days, for a total of 6 times between 10 February and 2 March 2014 and recorded the Universal Transverse Mercator coordinates for each sample location. While wearing clean latex gloves, we collected each scat sample found on hiking trails, placed it in a whirl-pak bag, and stored it in a freezer at -18 °C. We used razor blades to scrape epithelial cells from the outside of the scat and standard procedures to extract DNA (QIAmp DNA Stool Mini Kits, Qiagen, Los Angeles, CA); we included 1 negative control in each batch of extractions. To discern Coyotes from Vulpes vulpes L. (Red Fox) and Canis familiaris L. (Domestic Dog), we amplified a portion of the cytochrome-b region of mitochondrial DNA using the scatID primers IDF (5'-TATGCCTGATTCTACAGAT-3') and IDR (5'-TAGTATAGTCCTCGTCC-3') (Adams et al. 2003) in 20-µl reactions of 5 µl DNA extract, 0.5 pmol of each primer, 1x reaction buffer, 1.5 mM MgCl<sub>2</sub>, 1.5 units Amplitaq polymerase (Roche, Besel, Switzerland), 0.2 mM dNTPs, and 1 mg/mL BSA. We conducted polymerase chain reactions (PCR) in thermocyclers programmed for an initial denaturation at 95 °C for 10 min, 35 cycles of 30 s at 95 °C, 30s at 52 °C, and 40s at 72 °C, with a final extension of 72 °C for 3 minutes. Each PCR contained one blank control as well as the negative controls from DNA extractions. We digested PCR products with a restriction enzyme (Prugh and Ritland 2005) Taq<sup>a</sup> 1 (New England Biolabs Inc., Beverly, MA) overnight in a 37 °C water-bath and visualized the resulting fragments on agarose gels. Coyotes have band lengths of 100 bp, while other carnivores (e.g., Fox, Dog, Wolf) possess 200bp fragments (Kays et al. 2008).

We subsequently identified individual Coyote samples via multilocus genotyping with the microsatellites FH2001, FH2062, and FH2140 (Kays et al. 2008, Kohn et al. 1999, Mellersh et al. 1997) and labeled each forward primer with the following fluorescent dyes: 6-FAM for 2001, HEX for 2062, and NED for 2140. We performed PCR by running each microsatellite marker individually in 25-µl reactions that contained 2.0 µl of DNA extract, along with 1x buffer (Qiagen), 0.2 mmol dNTP, 1.5 mmol MgCl<sub>2</sub>, 0.5 µmol of the appropriate primer, 0.02 U/µL Taq polymerase (Qiagen), and TaqStart antibody (Invitrogen, Carlsbad, CA) with 14:1 antibody: Taq molar ratio. For each primer, our thermocycling profile included a 95 °C initial cycle for 3 min, followed by 40 cycles of 94 °C for 30 s, 58 °C for 30 s (60 °C for the 2062 primer), 72 °C for 45 s, and a final cycle of 72 °C for 2 min. We ran negative controls with each primer, and performed fragment-size analysis using a capillary automated DNA sequencer and Genemapper software (Applied Biosystems, Thermo Fisher Scientific, Waltham, MA). After all microsatellites were run 3 times, we calculated the probability of identification (Waits et al. 2001) and used GENEPOP (Rousset 2008) to determine whether the population was in Hardy-Weinberg equilibrium and to check for linkage disequilibrium.

For each individual Coyote, we constructed a capture history based on the resulting microsatellite identifications. Using program MARK, we developed and tested several a priori Huggins closed-capture models (White 2008) to determine population size and account for capture probability varying by a sampling effect (i.e., that initial capture probability would be greater than subsequent recapture probability because trails were not cleared prior to our first sampling occasion), capture period, and weather conditions. The weather variables that we identified as potentially important included snowfall ( $\Delta$ cm), time since last snowfall (hr), and density of snow cover (kg/m<sup>3</sup>), and we collected these data from the ARBW3 and Y01W3 weather stations in Madison, WI. We ranked our population models using Akaike's information criterion (AIC) (Burnham et al. 2011).

To quantify Coyote diets, we dried ½ of each scat sample at 90 °C for 48 h. We then washed scat samples with a size-60 strainer to separate large indigestible matter into bones, hair, feathers, and plant matter. We compared medulla patterning of hair samples (Moore 1974) to voucher specimens to identify mammalian species. Coyote diet-classification categories included *Odocoileus virginianus* Zimmermann (White-tailed Deer), *Sylvilagus floridanus* J.A. Allen (Eastern Cottontail), small mammals (including *Microtus* spp. [voles], *Peromyscus* spp. [mice], *Blarina brevicauda* Say [Northern Short-tailed Shrew], *Sorex* spp. [shrews]), *Sciurus carolensis* Gmelin (Gray Squirrel), birds, and vegetation. For scats containing more than 1 type of dietary item, we visually quantified the proportion of each item in each individual scat (Lukasik and Alexander 2012). We then calculated the relative frequency of prey items in all samples (Lukasik and Alexander 2012). Additionally, we calculated Shannon-Wiener diversity for the prey items in each Coyote scat sample (Krebs 1999).

We calculated habitat availability as the proportion of each of the 6 habitat types that occurred within a 1-m buffer along the length of the trails we sampled. We quantified habitat use by recording the habitat from which we collected each scat sample. We used Manly's alpha preference index to test for habitat selection (Krebs 1999) and inferred selection for a particular habitat type if the observed index exceeded the expected value.

#### Results

We collected 44 samples that we identified as Coyote scat in the field; 17 of these were eliminated after a second visual assessment in the lab (Halfpenny 2006). All 27 of the samples that we genetically analyzed for species identification were determined to be from Coyotes. Of these samples, 19 yielded complete genotypes, 7 amplified at 2 microsatellites, and 1 failed to amplify. From these genotypes, we identified 10 individuals with  $P_{\rm ID} = 0.004$  for samples with complete genotypes (n = 19) and  $P_{\rm ID} = 0.043$  for samples that amplified at microsatellites 2001 and 2062 (n = 7). We employed locus-specific tests to detect departures from HWE at locus 2140 (P = 0.014); tests for genotypic linkage disequilibrium were not statistically significant (P > 0.09). We found strong support for our top-ranking model, which accounted for a sampling effect, with the first sampling session featuring a greater capture probability ( $P_1 = 0.60 \pm 0.17$ ;  $\pm 1$  SE) compared to capture and recapture probabilities in subsequent sessions ( $P_{2-5}$ ,  $c = 0.19 \pm 0.06$ ). This model yielded a derived population estimate of 10–19 Coyotes (95% CI; Table 1).

#### Urban Naturalist T. Garwood, R. Ziobro, K. Keene, A. Waller, and J.N. Pauli

Of the scats analyzed for diet (n = 27), we found that Eastern Cottontails and vegetation were the most common dietary items, followed by Gray Squirrels and White-tailed Deer. Small mammals and birds were less frequent (Table 2). We did not detect any evidence of domestic animals. Prey types ( $H = 1.29 \pm 0.21$ ) were consumed relatively evenly ( $H_E = 0.66 \pm 0.11$ ).

We eliminated 3 of the Coyote scats used in the dietary analysis from habitatselection analysis because they were collected on trails other than those which we specified as sampling areas for this study. We found Coyote scat (n = 24), in 4 of the 6 sampled habitat types, with the majority being found in horticultural habitat (n =10), which comprised the smallest proportion of the sampled area (0.05%). Selection indices suggested that Coyotes strongly selected for the horticulture-cover type compared to all other habitat types (Table 3).

Table 1. Estimated Coyote population size (n) and 95% confidence interval ranked by AIC<sub>c</sub> in the University of Wisconsin-Madison Arboretum from a priori models.

Model	AIC <sub>c</sub>	ΔAIC <sub>c</sub>	W <sub>i</sub>	Model likelihood	Parameter	n	95%CI
Sampling effect $(P_1, P_{2-5} = c)$	66.8	0	0.83	1	2	11.6	10.3–19.4
Constant $P(P = C)$	72.2	0.62	0.24	0.07	1	12.3	10.5-21.5
Time varying $P(P_t = C_t)$	72.4	0.76	0.22	0.06	6	11.5	10.2-19.1
Change in Snow depth $(P_{\Delta S} C_{\Delta S})$	73.3	1.68	0.14	0.04	4	10.3	10.0-14.2
Snow density $(P_{\text{density}} C_{\text{density}})$	75.0	3.41	0.06	0.02	4	10.4	10.0-15.8

Table 2. Occurrence and average amount  $(\pm 1 \text{ SE})$  of each prey type within sampled Coyote scat as percentages. Occurrence denotes the proportion at which the prey item was present in all scat and amount denotes the proportion of the prey item in the individual scats, when present.

Diet item	Occurrence	Amount	
Eastern Cottontail	0.593	0.492 (0.089)	
Gray Squirrel	0.185	0.108 (0.053)	
White-tailed Deer	0.220	0.134 (0.058)	
Small mammal	0.074	0.050 (0.039)	
Bird	0.042	0.004 (0.004)	
Vegetation	0.296	0.173 (0.066)	

Table 3. Availability, use, and selection (Manly's  $\alpha$ ) of habitat type by Coyotes in the University of Wisconsin-Madison Arboretum;  $\alpha > 0.167$  indicates selection for that habitat. Coyotes selected strongly for the horticulture cover type.

Habitat	Availability	Use	α
Coniferous forest	0.160	0.083	0.006
Deciduous forest	0.369	0.208	0.007
Savanna	0.062	0.000	0.000
Prairie	0.390	0.292	0.009
Horticulture	0.005	0.417	0.978
Wetlands	0.013	0.000	0.000

5

2015

No. 5

#### Discussion

Using genetic-based approaches, we obtained an estimate of Coyote population size that indicated that the species is relatively abundant within the University of Wisconsin-Madison Arboretum. Our estimate of 10–19 Coyotes in the park (or approximately 0.02 Coyotes/ha) was comparable to the densities reported by Gehrt (2004) and Gese et al. (2012) in the Chicago area. Coyotes began recolonizing the Madison area in the latter half of the 20<sup>th</sup> century after nearly a hundred years of rarity (Schorger 1973). This expansion of the species into an urban area in southern Wisconsin follows a more general trend of predator repatriation across urban land-scapes (Bateman and Fleming 2012, Kertson et al. 2011).

Although Covotes appear to be relatively abundant in the arboretum, they still appear to be primarily subsisting on native prey. We found that Eastern Cottontail, Gray Squirrel, and White-tailed Deer were the most important dietary items, and found no evidence of pets in Coyote diets. The lack of pet remains in our samples may allay fears that a high density of Coyotes in urban areas leads to increased pet predation. However, because pets are likely kept inside more often during the winter, our work might underestimate Coyote depredation of domestic pets. This is especially relevant because the winter of 2013-2014 was unusually severe with low temperatures and deep snow. It would be interesting to compare the diet of Coyotes across years and seasons to better understand their use of native versus domestic prey. In terms of diversity of diet, our calculated dietary-diversity index is similar to those calculated for Coyotes in other human-altered landscapes, and is lower than those for Coyotes inhabiting natural areas (Kozlowski et al. 2008, Lukasik and Alexander 2012). A possible explanation for lower diet diversity in urban areas is that Coyotes prey on the other syanthropic wildlife species, which are abundant and potentially more available and vulnerable in an urban setting.

Unexpectedly, Coyotes selected strongly only for horticultural areas and avoided all other cover types. We originally hypothesized that Coyotes would select forest and prairie cover types because of the enhanced vertical cover, and that they would select against horticulture because of increased encounters with humans (Gese et al. 2012). It seems likely that Coyotes avoided humans by traveling at night (Gese et al. 2012, Grubbs and Krausman 2009), rather than by selecting for habitats with greater cover. Further, the risk associated with human encounters might not be as high as it might have been in other urban locations because dogs and firearms are not permitted in our study site. It is also possible that using scats to estimate habitat selection could introduce an unknown source of bias compared to the actual time Coyotes spent in different habitat types (for example, use of foraging versus resting sites). Based on our observations of abundant primary prey—Eastern Cottontails and Gray Squirrels, in particular—inhabiting the horticulture cover type, we suspect that scat sampling results do accurately reflect habitat use, especially related to foraging.

Previous studies have found that Coyotes in urban landscapes select for remnants of natural habitats (Gese et al. 2012, Grubbs and Krausman 2009). We found that within a largely natural habitat, Coyotes selected for the only non-natural cover

No. 5

type present. The severity of the 2013–2014 winter may help to explain the strong selection for horticulture. Edible vegetation was scarce and snow cover made off-trail travel difficult in natural cover types; however, there is a high density of paths and fruit trees at this site that may attract prey species as well as Coyotes. Thus, the diversity of resources in the horticulture-cover type may explain our observation of higher Coyote habitat use.

Our noninvasive and genetic-based approach provided insight into the habitat selection, diet, and population size of Coyotes in an urban landscape, which can serve as a baseline for the University of Wisconsin-Madison Arboretum and other urban landscapes. Future work could explore the differences in Coyote diet, habitat use, and population size between summer and winter months to develop a more complete understanding of this recolonizing population of Coyotes.

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