Red foxes (*Vulpes vulpes*) and coyotes (*Canis latrans*) in an urban landscape: prevalence and risk factors for disease

Meghan Pluemer,¹,* Shelli Dubay,¹ David Drake,² Shawn Crimmins,¹ Tessa Veverka,¹ Holly Hovanec,²,³ Miranda Torkelson³ and Marcus Mueller²

¹College of Natural Resources, University of Wisconsin – Stevens Point, 800 Reserve Street, Stevens Point, WI 54481, USA, ²Department of Forest and Wildlife Ecology, University of Wisconsin – Madison, 1630 Linden Drive, Madison, WI 53706, USA and ³School of Veterinary Medicine, University of Wisconsin-Madison, 2005 Linden Drive, Madison, WI 53706, USA

*Corresponding author. E-mail: pluemer.meghan@gmail.com

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Abstract

Urbanized areas contain fragmented landscapes and abundant resources, resulting in concentrated and increased wildlife populations in relatively close contact with other wildlife species, humans, and their domestic pets, thereby posing novel disease risks and facilitating inter-specific disease transmission. We trapped and radio-collared 15 red foxes (*Vulpes vulpes*) and 14 coyotes (*Canis latrans*) in the urban landscape of Madison, Wisconsin, to determine the prevalence of disease among these canids and to examine how these canids were using the landscape. Using Fisher’s exact probability tests, we found that coyotes had a significantly higher seroprevalence of Lyme disease (*P* = 0.002) and a higher prevalence of canine heartworm disease (*P* = 0.02) than foxes. Red foxes did not select specific habitat types in the urban landscape, but coyotes selected for forest and grass cover types, and avoided developed sites. Understanding the prevalence of disease in urban canid populations is important because diseases affecting urban canids cause morbidity and mortality and are transmissible to domestic dogs, and vice versa. Additionally, urban canids may serve as sentinels for zoonotic diseases such as Lyme disease and leptospirosis.

Key words: red fox, coyote, land cover, Lyme disease, heartworm

Introduction

Urbanization has intensified in recent decades, with 85% of the human population in the United States living in urban landscapes (McCleery et al. 2014). In addition to a dense concentration of people in human-modified landscapes, dense concentrations of wildlife species share the same areas. Two such species are the red fox (*Vulpes vulpes*) and the coyote (*Canis latrans*). Both species exist in sizeable populations within and throughout most urban areas in North America (Gehrt et al. 2010; Bateman and Fleming 2012). Recent increases in urban canid populations have occurred because many canid species are generalists and can thrive by utilizing a variety of habitats and food resources, thus allowing them to colonize human-dominated environments (Gehrt et al. 2010; McCleery et al. 2014).

One consequence of urbanization is increased interactions between humans, domestic animals, and wildlife. Veterinarians,
wildlife managers, and the general public in urban areas are concerned about interactions between wild canids and domestic pets, including direct and indirect disease transmission across species (Grinder and Krausman 2001; Malmlov et al. 2014). Urban wildlife is in close proximity to humans, and therefore viewed as a risk factor for zoonotic disease transmission (McCleery et al. 2014). This is especially relevant considering over 60% of emerging infectious diseases are zoonotic, with over 75% of these originating in wildlife (Jones et al. 2008). Additionally, fragmentation of natural areas in urbanized landscapes and the pattern of use of these habitats by wildlife may lead to increased densities of vectors and hosts, possibly resulting in a higher prevalence of disease in urban areas (Bradley and Altizer 2007).

Most studies to determine the seroprevalence of diseases in red fox and coyotes have been conducted in rural areas, finding varying prevalence rates of canine distemper virus (CDV), canine adenovirus (CAV), canine heartworm, canine parvovirus (CPV), Lyme disease (borreliosis), and leptospirosis. Given this known disease portfolio, Akerstedt et al. (2010) suggested that wild canids should be viewed as potential sources of infection to unvaccinated dogs. Similarly, Frölich et al. (2000) suggested that free ranging foxes may become infected with certain diseases from domestic dogs. Additionally, several of the diseases carried by urban canids such as leptospirosis and Lyme disease can be directly or indirectly spread to humans (Leighton and Kuiken 2001).

Identifying and understanding the prevalence of disease in red foxes and coyotes is important because disease may cause significant morbidity and mortality in urban canid populations but has received relatively little research attention. Furthermore, understanding disease prevalence in wild canids in urban areas is especially important because these canids may act as disease reservoirs, increasing risk of infection for domestic pets, and because domestic pets might transmit disease to urban canids. Urban canids also can be used as sentinels to predict the prevalence of zoonotic diseases such as Lyme disease and leptospirosis. Therefore, the objectives of our study were as follows: (i) identify disease exposure in urban red foxes and coyotes and (ii) determine if specific land cover characteristics and the presence of domestic dogs could explain the presence or absence of select diseases in these wild canids.

We hypothesized that disease prevalence and exposure would be higher for canids that spent more time in land covers conducive to disease transmission and with increased potential interactions with domestic dogs.

**Methods**

**Study area**

We studied red foxes and coyotes in Madison, WI, an urban area in Dane County (Fig. 1). Madison is the second largest city in Wisconsin, with a human population of 252,551 (U.S. Census Bureau 2016). Mean temperatures ranged from −10.4°C in winter to 20.6°C in summer with mean yearly precipitation of 873.8 mm (http://www.aos.wisc.edu/~sco/climhistory/state/4700-climo.html). Our 7102-ha study area encompassed the University of Wisconsin-Madison (UW) campus, along with a mosaic of residential and commercial areas bounded by developed roads. The study area also contained several semi-isolated, public natural areas, including the UW Lakeshore Nature Preserve (121.41 ha), UW Arboretum (485.63 ha), and Owen Conservation Park (39.25 ha). These natural areas consisted of upland broadleaf deciduous forests (oak [Quercus spp.], hickory [Carya spp.], ash [Fraxinus spp.], basswood [Tilia americana]), restored tallgrass prairies [indiangrass [Sorghastrum nutans], big bluestem (Andropogon gerardii], little bluestem (Schizachyrium scoparium)], and various wetland complexes [cat-tail (Typha spp.)] (Mueller 2017). Wooded corridors existed throughout the study area.

**Capture and monitoring**

Using cable restraints, we live-captured red foxes and adult coyotes from January 2015 to April 2018. For full capture details, see Mueller et al. (2018). All ethical capture procedures and trapping regulations for cable restraints were followed (Association of Fish and Wildlife Agencies 2017), and all animal handling methods were approved by the University of Wisconsin Animal Care Use Committee (Protocol A01559), and the Wisconsin Department of Natural Resources (WDNR) (Permit # SCP-SOD-001-2014).

We fitted each animal with ear tags and a very high frequency (VHF) radio collar (Advanced Telemetry Systems, Isanti, MN; Model M1950 for red fox and M2220B for coyote) or Global Positioning System (GPS) collar (Lotek Wireless Fish & Wildlife Monitoring, Newmarket, ON; Model G5C175C). Once an animal was radio-collared and released, it was located 2–3 times within the first 5 days to ensure it was moving and alive. Following the initial 5-day period, each VHF-collared animal was located at least once per week for the entire duration that the radio collar functioned and remained on the animal. Each location was triangulated based on the intersections of at least three telemetry bearings taken within a maximum of 15 minutes of each other to reduce error based on animal movement (Schmutz and White 1990). Animals also were located using Global Positioning System readings if individuals were visually observed. To ensure the accuracy of triangulations, telemetry bearings were plotted and the estimated location of the animal was shown on a laptop computer to proof locations in the field (unpublished data, Radio-Tracker, John Cary, University of Wisconsin, Madison, WI). During the weekly location of each animal, each animal was tracked for a 5-hour period, where it was located once per hour during that period. Weekly tracking periods were systematically rotated around the 24-hour clock to ensure that temporal variation in activity was captured. All GPS collars collected locations at hourly intervals and followed the same data collection schedule as VHF radio collars.

**Sample collection and processing**

For each trapped canid, we collected up to 6.0 ml of blood from the cephalic vein. We placed 1.5 ml in an EDTA tube and the remainder in a serum separator tube. Testing was completed at the University of Wisconsin-Madison School of Veterinary Medicine Clinical Pathology Laboratory, the Companion Animal Vaccine and Immuno Diagnostic Service Laboratory (CAVIDS), and the Wisconsin Veterinary Diagnostic Laboratory (WVDL).

**Vector-borne disease screening**

We used the SNAP 4Dx Plus (IDEXX Laboratories, Westbrook, ME) test, a rapid qualitative ELISA test that uses EDTA whole blood, to simultaneously detect heartworm (Dirofilaria immitis) antigen and antibodies against Lyme disease (Borrelia burgdorferi), Anaplasma phagocytophilum, A. platys, Ehrlichia canis and E. ewingii (Bowman et al. 2009). Additionally, Dirofilaria immitis microfilariae were noted when visualized in manual slide examination.
Serology

Samples with limited serum were first screened with the Canine VacciCheck kit (Spectrum Labs, Phoenix, AZ), a semi-quantitative modified ELISA enzyme labeled assay for CAV, CDV, and CPV. This assay measures on a scale from 0 to 6, with three set to a protective antibody titer of 1:16 for CAV, 1:32 for CDV, and 1:80 for CPV (Mazar et al. 2009). If samples had any result greater than 0 on this assay, the sample proceeded to undergo a gold standard quantitative confirmatory test, as described below. Based on crude data from a field and experimental trial of this test compared to gold standard quantitative confirmatory tests, using a score of 1 or greater as positive for exposure and using our titer cutoffs of $\geq 1:16$ for CDV and $\geq 1:20$ for CAV and CPV, there were very few false-negatives with sensitivity of 99.2% (CDV), 98.2% (CAV), and 99.4% (CPV) (Mazar et al. 2009).

Hemagglutination inhibition assay for CPV 2 antibody

We washed porcine red blood cells prepared in Alsever’s solution (Sigma A3551) four times with a buffer of 2M NaH₂P0₄ and 9% NaCl at pH of 6.8 prepared in Milli-Q water. Packed red blood cells were placed in buffer with 0.1% Bovine Serum Albumin (Sigma A9647) to make a 1% pRBC solution. Sera were 2-fold serially diluted in a 96-well plate with buffer. The prepared 1% pRBCs were added to all wells, and the assay was incubated at 4°C for 4–24 hours. The dilution of the last well where hemagglutination was inhibited was the reported titer. Duplicate positive and negative controls were run along with samples. We considered a titer of $\geq 1:20$ for parvovirus to indicate previous exposure (see Table 2).

Serum virus neutralization for CAV 1 and 2 and CDV antibody

Serum samples were 2-fold serially diluted in a 96-well plate with appropriate cell culture medium (CAV:DMEM, CDV:MEM). A control plate with positive and negative reference sera were run concurrently. The appropriate viral stock was diluted to 100 TCID50 and added to all test and control sera wells. A Viral Backtiter was placed on the control plate by serially diluting virus with culture medium as a check for appropriate dilution of stock. The remaining rows of the control plate were cell control, without added virus. The plates were incubated for 1 hour at 37°C. Cells from appropriate cell lines (CAV:MDCK, CDV:VERO) were added to control and test plates. Plates were incubated at 37°C 5% CO₂ for 4–5 days and were examined for the presence of cytopathic effects. The antibody titer was the highest dilution of serum that neutralized the virus and prevented infection of cells and resulting cytopathic effects. Titers $\geq 1:20$ were considered seropositive for CAV, and $\geq 1:16$ was considered seropositive for CDV (see Table 2).

6-Serovar microscopic agglutination test for Leptospira

The microscopic agglutination test for the detection of Leptospira antibodies in animal sera was run at WVDL using a protocol developed and distributed by the National Veterinary Services Laboratory (NVSL) (NVSL 1987). This assay undergoes annual proficiency testing distributed by the World Organization for Animal Health (OIE). The serovars tested are six most commonly found within canids in the midwestern United States: Leptospira interrogans bratislava, L. i. icterohemorrhagiae, L. i. canicola, L. i. pomona, L. i. grippotyphosa, L. i. autumnalis. Titer results $\geq 1:100$ were interpreted as consistent with current or past exposure based on previous research (Table 1), with titers $\geq 1:1600$ suggestive of recent infection based on currently accepted cutoffs for a single titer in domestic dogs (Miller et al. 2011).

Spatial data variables

Geographic Information System (GIS; ESRI 2017. ArcGIS Desktop: Release 10.4. Environmental Systems Research Institute, Redlands, CA) was used to create a map of the study area based on National Land Cover Data (Homer et al. 2015). The NLCD...
types were aggregated into four categories: water (i.e. open bodies of water, woody wetlands, and emergent herbaceous wetlands), forest (i.e. deciduous, evergreen, mixed forest, and shrub and scrub), developed (i.e. lawn grasses and impervious surfaces accounting for 20–100% of cover), and non-woody undeveloped land cover (i.e. grassland and herbaceous, pasture and hay, and cultivated crops, hereafter referred to as grass).

We contacted the City of Madison Treasurer’s Office and received the records of domestic dogs in the city of Madison, Wisconsin, registered from 2013 to 2016. The location of each household with a reported registered dog within our study area was digitized into ArcMap 10.4.

Statistical analysis

We used a Fisher’s exact probability test to determine if exposure to CPV, CDV, CAV, Lyme borreliosis, and leptospirosis varied significantly between species. We also used a Fisher’s exact test to determine if infection with heartworm differed by species.

We used two approaches to assess the potential impacts of habitat selection on disease exposure. First, we calculated selection ratios for each aggregated land cover type in the study area (McDonald et al. 2012). We compared the proportion of the study area in each aggregated land cover type to the proportion of telemetry locations within each land cover type for each animal. We used a t-test to determine if selection ratios differed significantly from 1 (McDonald et al. 2012). We calculated parameter estimates and p-values from resource selection functions for each species (Mueller 2017). We did not include telemetry data gathered during the winter months (November–February) as we assumed during these periods that the active vectors for disease transmission (mosquitoes and ticks) were inactive. We further limited our analyses to animals for which we had at least 10 locations.

Results

Prevalence of disease

We captured and radio-collared 15 red foxes (10 males and 5 females) and 14 coyotes (7 males and 7 female) in our study area. Of these canids, none of the foxes were infected with heartworm, while five (35.7%) coyotes were infected. Only one (6.7%) fox had exposure to Lyme borreliosis. Nine of the fourteen (64.3%) radio-collared coyotes tested positive for Lyme exposure. All 14 coyotes and 15 foxes were seronegative for ehrlichiosis and only 1 of the 15 (6.6%) foxes was seropositive for Anaplasma antibody. Not all samples taken from the canids at the time of their capture contained a sufficient amount of blood to run all of the disease tests, so not all canids were able to be tested for exposure to parvovirus, adenovirus, distemper, or leptospirosis. Of the 15 foxes and 11 coyotes that were tested for CPV antibody, 8 foxes (53.3%) and 5 coyotes (45.5%) tested positive. For CAV, 1 of 10 (10%) coyotes and 2 of 14 (14.3%) foxes were seropositive. Three of eleven (27.3%) coyotes and two of thirteen (13.3%) foxes were seropositive for CDV antibody. Lastly, 1 of 14 (7.1%) coyotes and two of fifteen (13.3%) foxes had exposure to leptospirosis, with one fox having a titer of 1:3200 against L. i. grippotyphosa and 1:1600 against L. i. autumnalis, titers consistent with active or recent infection.

Exposure to CPV ($P = 0.50$), CAV ($P = 0.63$), CDV ($P = 0.35$), and leptospirosis ($P = 0.53$) did not differ between the two species. However, coyotes had a higher prevalence of both Lyme antibody ($P = 0.002$) and canine heartworm antigen ($P = 0.02$) than red foxes.

Habitat selection

Red foxes appeared to exhibit random selection for land cover across the study area, with no selection ratios differing significantly from 1 (Fig. 2). Coyotes exhibited significant selection for both forest and grass cover types and avoided developed areas (Fig. 2). When including the distribution of domestic dogs and accounting for animal movements, we found that foxes exhibited random selection of all cover types while coyotes exhibited moderate ($P = 0.068$) avoidance of developed cover types (Table 1).

Discussion

Canine heartworm

Canine heartworm is a mosquito-borne disease caused by the nematode *Dirofilaria immitis*. Pulmonary and respiratory function can be affected when the parasites inhabit the heart and arteries of the canid (Anderson 2001; Nelson et al. 2003). Thirty-six percent of the coyotes trapped in Madison, WI, were infected with heartworm, and heartworm in coyotes was significantly greater than in red foxes (0%). The abundance of the parasite may differ by physical location, amount of precipitation, and density of mosquito vectors (Wixson et al. 1991). Previous studies have reported that canids living near wooded wetlands and rivers have the highest prevalence of heartworm (Pappas and Lunzman 1985; Gortazar et al. 1994; Nelson et al. 2003). Holzman

Table 1: Parameter estimates and P-values from resource selection functions for red foxes and coyotes in Madison, WI, 2015–2018

|            | $\beta$  | SE     | p    |
|------------|----------|--------|------|
| Coyote     |          |        |      |
| Intercept  | -0.00033 | 0.03800| 0.993|
| Water      | -0.08940 | 0.10459| 0.393|
| Forest     | 0.06446  | 0.07452| 0.387|
| Developed  | -0.15251 | 0.08364| 0.068|
| Grass      | 0.03931  | 0.05598| 0.483|
| Dogs       | -0.02338 | 0.04964| 0.638|
| Fox        |          |        |      |
| Intercept  | 0.00001  | 0.00582| 0.909|
| Water      | -0.01400 | 0.00864| 0.105|
| Forest     | -0.00082 | 0.00768| 0.915|
| Developed  | -0.01522 | 0.00996| 0.126|
| Grass      | 0.00256  | 0.00653| 0.695|
| Dogs       | 0.00918  | 0.00670| 0.171|


Perhaps screening the blood of foxes for both microfilariae and ducce microfilaria which then can be transmitted to foxes. nid hosts, such as coyotes and domestic dogs, to infect and pro-

and Youatt (1972) speculated that the parasite requires other ca-

Wixsom et al. (1991). Why prevalence differs between canid spe-

heartworm infection compared to coyotes or domestic dogs

Wixsom et al. (1991). Overall, foxes tend to show lower levels

for the lack of heartworm in coyotes. We found that none of the foxes in our study were infected

with heartworm. The trend of heartworm being less prevalent

in foxes has been found in other studies as well (Stuht and

and Youatt 1972; Pappas and Lunzman 1985). Stuht

with heartworm. The trend of heartworm being less prevalent

as a sentinel species (Lindenmayer et al. 1991; Olson et al. 2000).

Olson et al. (2000) found coyotes to have lower seropositivity

than domestic dogs, despite coyotes seeming to have a greater

risk of exposure to Lyme disease.

The lower prevalence of Lyme disease in red foxes may be
due to foxes being resistant to the bacterium. Heidrich et al.
(1999) concluded that the red fox is a minor reservoir for the
bacterium because research in Germany showed that only
seven of 100 red fox skin samples tested positive for B. burgdor-
feri. Dumitrache et al. (2015) and Kahl and Geue (1998) posited
that red foxes likely play a minor role in the persistence of Lyme
disease.

Canine parvovirus

CPV is a highly contagious viral disease that was first reported
in domestic dogs in the late 1970s (Goddard and Leisewitz 2010;
Zourkas et al. 2015). Transmission among wild canids and do-
mestic dogs is facilitated because CPV is long-lasting in the en-

vironment and is easily acquired through direct contact with
contaminated feces, urine, and other bodily secretions or
through contact with infected fomites or environments
(Osterhaus et al. 1980; Zourkas et al. 2015). We found that
approximately 50% of the coyotes and foxes tested were exposed
to CPV, although many other studies have identified much
higher prevalences in the United States (Table 2). Canuti et al.
(2017) found that CPV was more prevalent in coyotes than in
foxes; however, we found no difference in exposure by species.
Transmission may also increase in urban areas through in-
creased contact between domestic dogs and other urban wild-
life (Truyen et al. 1998). However, wild canid-domestic dog
interaction was not a significant variable in any of our models,
so it is likely that contact between wild canids and domestic
dogs is uncommon within our study area, perhaps because do-
mestic dogs in Madison, WI, are vaccinated, thus decreasing
the risk for urban wildlife to contract parvovirus from indirect or
direct contact with domestic dogs.

Leptospirosis

Leptospira interrogans is a bacterial pathogen that is most com-
monly transmitted to wildlife and humans through contami-
nated drinking water (Leighton and Kuiken 2001). We found low
prevalence of exposure to leptospirosis in red foxes (7%) and
coyotes (13%). Prevalence was similar to many other studies
(Table 2), but lower than that reported by Amundson and Yuill
(1981) for red foxes (47%) trapped in southwestern Wisconsin.
Amundson and Yuill (1981) showed that juvenile foxes were
more commonly exposed, potentially explaining why our results showed lower exposure in adult foxes. The infective titer documented in one red fox indicates that red foxes in urban environments may serve as risk factors for human leptospirosis since foxes regularly used developed areas in Madison, WI.

**Canine distemper**

CDV is transmitted primarily through contact with oral, respiratory, or ocular fluids, and the infective virus can be shed up to 90 days after exposure (Williams 2001). We found relatively low exposure to CDV in urban red foxes (13%) and coyotes (27%), and prevalence did not vary significantly with species. Prevalence for red foxes was similar to that reported in previous studies in Wisconsin (Table 2, Amundson and Yuill 1981). Prevalence in coyotes was similar to previous studies of urban coyotes (Table 2, Grinder and Krausman 2001). Low levels of canid exposure to CDV may be explained by the widespread, routine vaccination of domestic dogs against the virus.

**Canine adenovirus**

CAV, also known as infectious canine hepatitis, is caused by a double-stranded DNA virus and is transmitted via urine, nasal and conjunctival secretions, and feces (Woods 2001). Prevalence for CAV was low in coyotes (10%) and red foxes (14%) in our study. In the western United States, prevalences above 50% are common in adult coyotes (Table 2, Cypher et al. 1998). Only 3% of red foxes tested positive for CAV antibody in Wisconsin (Amundson and Yuill 1981), and our results reflect similar lower prevalence rates. In infected foxes, the virus may be transmitted among unvaccinated domestic dogs and wild canids (Woods 2001).
Perhaps coyotes and red foxes in Madison are not exposed at a higher rate because domestic dogs are vaccinated, decreasing the likelihood of coming into contact with an infectious host. Moreover, our models suggested that potential wild canid-domestic dog interactions were not a significant factor in our study area.

Comparison to other studies
In general, coyotes and foxes in Madison, WI, were not exposed to many of the pathogens we evaluated. When compared to other urban canids, seroprevalence was lower for CAV and CPV (Table 2). Coyotes trapped in Madison had similar exposure to CDV as coyotes trapped in Tucson, AZ, and red fox showed similar seroprevalence to CDV as foxes sampled previously in Wisconsin and in Norway (Table 2). Canids trapped in Madison, WI, were not exposed to L. interrogans as often as foxes sampled in Wisconsin previously (Table 2). Coyotes sampled in Madison, WI, were similar to rural Texas coyotes in exposure to Lyme borreliosis (Table 2).

The disease prevalences found in the urban canids in Madison, WI, were not always similar to other urban canid disease prevalences and, in some situations, more closely aligned with prevalences found in rural environments. Madison, WI, may not be a typical urban environment because urban green space is relatively common throughout the city. In 2016, 13.4% of Madison’s adjusted city area was parkland (Harnick et al. 2017). In comparison, Milwaukee, WI (the largest city in Wisconsin) had 10.3% parkland and Tucson, AZ, had 3.2% parkland in the adjusted city area (Harnick et al. 2017). The 100 largest urban cities in the United States averaged 3.7 parks per 10,000 residents in 2016, but Madison, WI, has about 11.6 parks per 10,000 residents (Harnick et al. 2017). Additionally, Madison, WI, hosts three relatively large natural areas (the UW Arboretum, UW Lakeshore Nature Preserve, and the Owen Conservation Park) which add considerable acreage of contiguous green space in comparison to other urban cities. Disease prevalences found in our study may be similar to disease prevalences in rural coyotes because many of our coyotes were trapped in and remained near the conservation parks and used the urban green spaces in Madison, WI. These urban green spaces may mimic rural areas in that perhaps they have similar mosquito or tick abundances and these urban green spaces likely serve as transmission sites for vector-borne diseases amongst the developed land cover of an urbanized landscape.

Habitat relationships with disease exposure
We hypothesized that disease prevalence and exposure would be higher for canids that spent more time in land covers conducive to disease transmission. The spatial partitioning employed by our study species seemed to affect the disease risk for urban coyotes and red foxes. We found radio-tracked coyotes in the Madison area selected for natural areas and avoided areas with moderate to high development. The habitat selection exhibited by these coyotes may help explain why coyotes had a higher prevalence of vector-borne diseases (canine heartworm and Lyme disease). The natural areas that coyotes selected for overlapped with habitat conducive to supporting the tick and mosquito vectors for these diseases. Additionally, vectors may become more concentrated in the fragmented natural areas of urban landscapes (Bradley and Altizer 2007). Tick abundance has been found to be positively associated with deciduous forests and negatively associated with grasslands (Guerra et al. 2002), but in an urbanized landscape, forest and grassland land cover are often fragmented. The fragmentation of these habitats may cause these areas to have high densities of ticks and an abundance of white-footed mice (Peromyscus leucopus), which are reservoirs for Lyme disease (Bradley and Altizer 2007), thereby resulting in a higher prevalence of Lyme disease in urban areas (Bradley and Altizer 2007). For example, five out of the six (83%) coyotes trapped in the UW-Madison Arboretum, a 485-hectare natural area consisting of wooded wetlands, deciduous forest, and grassland habitats, tested positive for heartworm antigen and Lyme antibody. Tick abundance has been found to be associated with forested land cover (Guerra et al. 2002; Wood and Lafferty 2013), and, in our study, coyotes were found to use natural areas more than the red foxes in the study area (Mueller 2017).

In contrast, red foxes selected for areas with low development and avoided natural areas. More developed areas (where red foxes were commonly found and where humans live and work) are typically treated with pesticides and offer less suitable habitat for ticks and mosquitoes due to mowed vegetation and less standing water, rendering these sites inadequate habitat for vectors.

Contrary to our hypothesis that disease prevalence and exposure in urban coyotes and red foxes would increase with increased potential interaction with domestic dogs, we did not find a relationship between potential contact with domestic dogs and home ranges of urban canids. First, domestic dogs tend to be more active during the day and reside inside at night, whereas urban coyotes and foxes are active at night and to a lesser degree around crepuscular hours, possibly limiting physical proximity between dogs and other canids. Second, coyotes tend to use urban green spaces, such as UW-Arboretum and Owen Park, while foxes more often use developed sites. We would expect that foxes would interact more often with domestic dogs, but our analysis did not render domestic dog contact as an important factor.

Conclusions
When compared to coyotes and foxes studied in other locations, urban red foxes and coyotes in Madison appear to be in relatively good health (Table 2). Coyotes, which selected for urban forest and grassland habitats, had higher seroprevalence of Lyme and higher infections of heartworm than red foxes. Both of these diseases are vector-borne, and the habitats used by coyotes are adequate habitat for ticks and mosquitoes as well. The developed areas more often used by foxes are likely treated for mosquitoes and ticks either by municipal governments or by homeowners. Additionally, red foxes may be poor definitive hosts for heartworm, and so coyotes are more likely to influence the transmission cycle in Madison. Overall, coyotes in urban Madison were found to have a low pathogen load. Coyotes were more likely to be exposed to Lyme disease, therefore, they may contribute to the natural Lyme disease transmission cycle in urban areas. However, coyotes were less likely to inhabit developed urban environments, so domestic animals and humans have relatively limited opportunity for interaction with coyotes in Madison. Red fox exposure to Leptospira interrogans could pose possible threats to domestic animal and human health because they select for urban developed habitats in closer proximity to people and their pets. Additionally, one fox had a titer indicative of recent exposure to L. interrogans. Further research into modes of leptospirosis and Lyme disease transmission among hosts in urban areas is warranted.
Data Availability
Data will be made available upon request.

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