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Community Variation of Gastrointestinal Parasites Found in Urban and Rural Coyotes (*Canis latrans*) of Calgary, Alberta

We compared parasite variation in coyote feces collected in eight urban sites within Calgary, Alberta, to those in six rural sites outside the city limits. Four hundred and sixty fresh fecal samples (< 4 days old) were collected weekly between July 2009 and June 2010. Fecal flotation analysis identified parasites to the level of genus. We calculated parasite richness, diversity, and evenness. Parasite richness was significantly greater in rural than urban sites. Disparities in the parasite community included a significant variation in parasite richness, diversity, and evenness among rural and urban sites. *Toxascaris leonina* and *Cystoisospora* spp. were shared among all urban sites, while these species in addition to *Taenia*-like spp. and *Trichuris* spp. were shared among all rural sites. Both urban and rural sites yielded evidence of *Toxocara canis*, *Taenia*-like spp., and *Giardia* spp., which are potential zoonotic parasites. *Toxocara canis* was at highest prevalence in a core urban site, Inglewood Wildlands, which exhibits unique biotic and abiotic characteristics. Factors of exposure to parasites, such as diet, behavior, and environmental factors, are suggested to be most influential on variation in urban and rural coyote parasitism. We suggest coyotes are an important focal species for further research in urban disease ecology, due to the resilient nature of the carnivore and its propensity to co-exist with people and domestic animals.

Keywords

coyotes, *canis latrans*, parasites, urban vs. rural, fecal analysis

INTRODUCTION

Urban environments can influence the ecology of wildlife disease systems, resulting in potential implications for public health (Bradley and Altizer 2006). While certain parasites are commonly found in most individuals of a given wildlife species, research has suggested that urban environments may increase parasite prevalence in wildlife species that live in cities (i.e., urban wildlife). For example, Leher et al. (2010) showed that the prevalence of Toxoplasmosis antibodies (causative agent: *Toxoplasma gondii*) in woodchucks (*Marmota marmax*) was positively related to urban land use in Champaign-Urbana, Illinois. Similarly, raccoons (*Procyon lotor*) in Southern Ontario, showed *Salmonella* prevalence from 27% in rural habitat versus 65% in urban habitat (Jardine et al. 2011). As urban areas exhibit high density residential, business, and industrial land-uses, these studies have postulated that certain wildlife species experience higher rates of parasitism and disease because of one or multiple of the following factors of susceptibility: increased stressors, poor nutrition, or increased frequency of inter- or intra-species interactions with other wild or domestic animals. However, exposure to parasites is the most important factor in parasite prevalence (Poulin 2006), and factors of susceptibility may facilitate infection only as a secondary result of exposure.

Exposure probability is limited by the transmission strategy of the parasitic organism and the presence or absence of that organism in an infective state, in a given habitat where hosts live. Wildlife species that move amongst habitat types may be frequently exposed to a greater number of parasite species. Understanding the differences in parasite assemblage of a terrestrial mammal with wide home-ranges can help illuminate potential exposure to parasites associated with using urban or rural environments. And, if that wide ranging species is one that comes in contact with humans or their pets it may be important to understanding health implications. Thus, as wide ranging (Atwood et al. 2004), top carnivores in the urban and rural environment, coyotes (*Canis latrans*) are likely an important focal species for such a comparative analysis.

Our work was predicated on concerns expressed in the media (Alexander and Quinn 2011) by the Canadian public, regarding the risk of disease transmission from coyotes to people or their pets. While most interactions with coyotes are benign (Alexander and Quinn 2011), studying parasitism in urban coyotes is important, as infected animals can pose infection risks for people and their pets (directly through physical contact or indirectly through fecal deposits) (Deplazes and Eckert 2001, Mani and Maguire 2009). Developing baseline data of the occurrence of parasites will also provided much needed information to dispel fears expressed by the public, or aid in management decision.

Urban coyotes in Canada are host to several viral pathogens including rabies, canine distemper virus, and canine adenovirus (Rosatte 1988, Grinder and Krausman 2001, Arjo et al. 2003), all of which can be health concerns to people or their pets. And, while the risk of human infection by viral coyote parasites (including rabies) is relatively low (Rosatte 1988), it remains an implication of coyotes co-existing with humans (Thompson et al. 2009). Public risks may also be presented by gastrointestinal parasites, many eggs of which are deposited in feces in core urban environments. For example, *Echinococcus multilocularis* and *E. granulosus*, are highly zoonotic parasitic species reported in foxes, wolves, and coyotes, especially in western North America. These specific parasites are zoonotic tapeworms that, in humans, can have an

asymptomatic incubation period of 5-15 years before a chronic symptomatic course, potentially resulting in death (Kern et al. 2003). While this is an extreme case of human risk, such parasites highlight the issue of health risks associated with wild canid parasites in public spaces.

Despite repeated reference by media sources about the disease risk posed by coyotes to Canadians (Alexander and Quinn 2011), we found no peer-reviewed, recent literature for Canadian cities that evaluate the occurrence of gastrointestinal parasites in coyote feces, nor the associated risks to the urban public. Surveys of coyote gastrointestinal parasitism were, however, found for more remote areas of Western North America since the 1960s (Holmes and Podesta 1968, Conder and Loveless 1978, Samuel et al. 1978, Seese et al. 1983, Storandt et al. 2002, Thompson et al. 2009). Many of these studies reported data from a single more remote area so the results were generally not transferrable to urban environments. Therefore, our current research addresses the noted deficiency of literature, but examined only the occurrence of parasites in coyote feces found in urban and rural environments. Our work represents a one year profile of parasites in coyote feces, and our principle aim was to provide baseline data on and characterize current coyote parasitism in Calgary, Alberta. A secondary aim was to contrast between parasite communities found in coyotes feces collected in urban or rural areas surrounding the city. We acknowledge that these data are temporally limited. Therefore, our results provide a baseline that is essential to building current understanding and for comparison in longitudinal studies, whereby variability through time and different climatic regimes could be exposed.

The parasite component community is defined here as all infra-populations of parasites associated with some subset of a) a host species, or b) a collection of free-living phases associated the external environment (Bush et al. 1997). By determining parasite richness, diversity, and evenness for the component community and comparing between urban and rural coyote estimates, we illuminated differences between these regions and assessed variability of parasitism within each region.

We defined richness as the number of species present in a collection, evenness is defined as a measure of disparity in the number of species identified in a collection (Bush et al. 1997), and diversity is the composition of a parasite community in terms of number of species present (Bush et al. 1997). These three measures are important in parasitology because landscape-scale differences can be elucidated, and we specifically chose these measures to identify general patterns of parasite prevalence across a large geographic region. Also, parasite richness, evenness, and diversity reflect the similarities and differences of parasites shared by coyote hosts among regions. Understanding the distributions of parasites can reveal patterns of parasite prevalence within given habitats and in comparison to other habitats of different habitat structure. Such differences could shed light on landscape- or patch-level biotic or abiotic factors that might influence exposure to these gastrointestinal parasites.

METHODS

Study Area

Calgary, Alberta is located at (51.083° N, 114.083° W) approximately 70 km from the Rocky Mountain front ranges and has a population density of 2,308 people per square kilometer of built-form area within the city limits (Calgary Land Use Policy and Planning 2010). Rural areas beyond the city limits have a population density of 6 people per square kilometer of agricultural land-area. Calgary offers an ideal biogeographic context for the measurement of urban wildlife parasitism, having a gradient of land uses extending from its urban core to rural areas. City park areas (i.e., greenspaces) are common within the city; while some are large enough to support coyotes, and most animals use both the greenspaces and the surrounding matrix of residential developments. Although no empirical measure of coyote abundance has taken place within Calgary, sighting reports and indirect evidence such as road kill carcasses, kill sites, and fecal samples indicate the presence of coyotes in most areas of the urban centre, including the core (Lukasik and Alexander 2010).

Beyond the city limits, the rural region is recognized as a “foothills” landscape, defined primarily by prairie environments divided as agricultural land-use. Farms are generally privately owned or leased government land, but some western Calgary rural areas also feature publically accessible natural or conservation areas. Coyotes are observed regularly in both private and public rural areas of Calgary.

Field Sampling

We used a non-invasive survey approach (Darimont et al. 2008), relying on the analysis of fecal samples from coyotes. Weekly searches for coyote fecal samples were conducted from July 2009 to June 2010 in eight urban sites and six rural sites described subsequently (Figs. 1 and 2). Coyotes are known to defecate on paths, trails, and road edges (Grubbs and Krausman 2009). Hence, searches for fecal samples were conducted along transects of at least 1 km in length, chosen by our ability to detect at least two fecal samples within this distance during preliminary investigation. A 1 m buffer from transects was used to cover entire paths or road edges where fecal samples were occasionally found. While fecal samples may have been deposited outside the transects, we assume that the fecal samples contained similar parasite species representative of the local coyote metapopulations in both urban and rural sites (Villanua et al. 2006).

Fecal samples were collected by two independent observers (one being the first author), who were experienced in coyote feces detection and trained in its identification by an experienced tracker, the second author. Fecal samples were identified by the following morphological characteristics typical of coyote scat: shape (elongated, tapered end), size (18mm to less than 25mm in diameter), texture (dense, firm, variable with age), colour (darker shade of brown or grey, occasionally roan), and deposition location (on path or road edges, as different from domestic canines) (Halfpenny and Biesiot 1986). Samples that satisfied these conditions were stored in sealed plastic bags, and placed in airtight bins for transportation from sites to a -20°C freezer for storage prior to laboratory analysis. We did a secondary visual screening of fecal samples and discarded any samples that raised questions of species origin.

Urban sites (Fig. 1) were chosen according to the following criteria: existence within city limits; known coyote presence (based on sightings reports and preliminary investigation); existence within one of four city quadrants; minimum 6 km distance from other urban sites (Gehrt et al. 2009); and recreational public use (Gibeau 1998). Urban sites included commonly used greenspaces and were consistent with those selected by Lukasik and Alexander (2010). Rural sites were chosen according to the following criteria: existence beyond city limits; accessibility – land ownership; high fecal sample recovery; and good representation of rural habitats. Rural sites (Fig. 2) corresponded to agricultural edges and roads or rural conservation area pathways. Rural sites were separated from urban sites by several average coyote home range diameters, and thus minimized the chance of cross contamination of samples by individual animals or contact amongst urban and rural animals.

Laboratory Analysis

All samples were frozen in an -80°C freezer for 72 hours prior to analysis to prevent the risk of investigator infection by potentially harmful parasite eggs, specifically *Echinococcus* spp. (Dryden et al. 2005). A double-centrifugation fecal flotation method was used to analyze coyote fecal samples (Zajac and Conboy 2006). This technique has been used for multiple surveys of coyote gastrointestinal parasitism in North America (Gau et al. 1999, Gompper et al. 2003, Lesmeister et al. 2008). Sheather's sugar solution with specific gravity 1.26 was mixed with 4g of fecal samples to facilitate parasite eggs and oocysts to rise to the top of the solution. The first author, who was trained in common gastrointestinal parasite diagnostics (Kutz Wildlife Parasitology Laboratory, University of Calgary), morphologically identified parasite eggs and oocysts on prepared slides for all samples. Identification of the parasites to genus, and when possible to species, was confirmed using keys of ova and cysts common in dog, wolf, coyote, and fox hosts (Bowman 2008). Morphological identification was assessed by parasite propagule size, shape, wall width, and distinctive arrangements or identity of internal components. Results were recorded as eggs/oocysts per gram feces for each parasite identified.

Statistical Analysis and Diversity Indices

Measures of the parasite component community (i.e., parasite richness, diversity, and evenness) were first computed in PRIMER v.6 2006 (Clarke and Gorky 2006) and then analyzed in SPSS v.19 (SPSS, 2010). Richness was quantified as the raw number of parasite taxa identified (S). Because some parasites were only confidently identified to the genus level, all levels of identifiable taxa were considered for species richness. Pielou's Index (J') was chosen to account for evenness, producing a value of heterogeneity in the relative representation of species present. Diversity was quantified using the Shannon Weiner Index (H'), which determines the proportion of individuals that represent each present species (S) (Magurran 2004). These non-parametric indices were selected due to their common use in ecology and application for statistical comparison of parasites (Magurran 2004), and diversity indices were chosen to identify patterns across sampling sites as well as between regions. Finally, a Student's t -tests for one-sample (variability among sites) and independent samples (variability between areas) were used for richness, diversity, and evenness, at $p < 0.05$.

Figure 1. Urban sites chosen for coyote fecal sampling in Calgary, Alberta.

Figure 2. Rural sites chosen for coyote fecal sampling outside the city limits of Calgary, Alberta.

RESULTS

Parasite Richness

Fifteen parasites were observed from 460 fecal samples from the Calgary region ($S = 15$). Parasites were identified to the genus level and included cestode, nematode, protozoa, and trematode species (Table 1). Some parasites were confidently identified to the species level. All parasites were observed in urban and rural areas, with the exception of *Paragonimus kellicotti*, which was found exclusively in rural sites.

TAXONOMIC GROUP	COMMON NAME	URBAN ($n=189$)		RURAL ($n=271$)		TOTAL ($n = 460$)	
		Prevalence (%)	Frequency	Prevalence (%)	Frequency	Prevalence (%)	Frequency
CESTODA		13.76	26	14.76	40	14.35	66
<i>Diphylobothrium</i> sp.	Broad Fish Tapeworm	0.5	1.0	0.7	2.0	0.65	3
<i>Dipylidium caninum</i>	Cucumber tapeworm	2.7	5.0	1.1	3.0	1.74	8
<i>Taenia</i> -like sp.	Tapeworm	10.6	20.0	13.3	36.0	12.17	56
NEMATODA		61.4	116.0	59.4	161	60.22	277
<i>Capillaria</i> sp.	Fox lungworm	2.1	4.0	6.3	17.0	4.57	21
Pinworm	Pinworm	0.5	1.0	0.4	1.0	0.43	2
<i>Physaloptera</i> sp.	Stomach worm	3.2	6.0	1.1	3.0	1.96	9
<i>Toxascaris leonina</i>	Roundworm	54.0	102.0	49.1	133.0	51.09	235
<i>Toxocara canis</i>	Dog roundworm	7.4	14.0	1.5	4.0	3.91	18
<i>Trichuris</i> sp.	Whipworm	4.8	9.0	12.2	33.0	9.13	42
<i>Uncinaria stenocephala</i>	Northern hookworm	4.2	8.0	1.5	4.0	2.61	12
PROTOZOA		13.8	26.0	9.6	26	11.30	52
<i>Cystoisospora</i> sp.	Coccidia	10.6	20.0	6.6	18.0	8.26	38
<i>Giardia</i> sp.	Giardia; Beaver Fever	0.5	1.0	0.4	1.0	0.43	2
<i>Sarcocystis</i> sp.	Sarcocystis	2.7	5.0	3.0	8.0	2.83	13
TREMATODA		1.6	3.0	3.0	8	2.39	11
<i>Alaria</i> sp.	Intestinal fluke	1.6	3.0	2.2	6.0	1.96	9
<i>Paragonimus kellicotti</i>	Lung fluke	-	-	0.7	2.0	0.43	2

Table 1. Parasite prevalence in urban and rural sites of Calgary, Alberta. Frequency is a measure of the number of samples tested positive for a given parasite species.

Parasite richness was significantly greater for rural sites ($S = 15$, $n = 271$) than urban sites ($S = 14$, $n = 189$) ($t = -3.308$, $df = 12$, $p = 0.006$), though the difference was due largely to the absence of *P. kellicotti* in any single urban sample (Table 1). Species richness was significantly different amongst urban sites ($t = 10.538$, $d.f. = 7$, p -value < 0.05). Similarly, species richness was significantly different amongst rural sites ($t = 19.852$, $d.f. = 5$, p -value < 0.05). In the urban sites, parasite richness ranged from 5 species (shared in richness by Arbor Lake, Stanley Park, Tom Campbell's Hill, Weaselhead Natural Area, and Fish Creek: Sicome Lake) to 9 species (recovered from the Inglewood Wildlands) (Fig 3). Inglewood had a high prevalence of *T. canis* and *Physaloptera* spp. relative to all other urban sites. In the rural sites, parasite species richness ranged from 7 species (recovered from Sandy Cross: Farm 5A) to 10 species (recovered from Big Hill Springs Provincial Park) (Fig. 4). In sum, from a total of fifteen parasites found, fourteen were shared between the urban and rural sites; although the mean value of richness for the rural region was greater than the urban region. Parasite richness was significantly different amongst sites within both urban and rural regions.

Figure 3. Parasite species richness of coyotes represented at each urban sampling site in Calgary, Alberta. Values within each graph represent the percent prevalence of the given parasite species from each site.

Figure 4. Parasite species richness of coyotes represented at each rural sampling site in Calgary, Alberta. Richness values are generally higher for all rural sites than those included in the previous map for urban sites.

Parasite Evenness

Pielou's Index was used as simple measure of evenness (J'). Urban and rural regions were not significantly different in parasite evenness ($t = 0.937$, d.f. = 1, $p = 0.367$) (Table 2). Evenness in the urban region was 0.671, which was very similar to the rural region evenness value of 0.647. Amongst all urban sites, the evenness of parasite species was significantly different ($t = 30.579$, d.f. = 7, $p < 0.05$). Evenness of parasites was also significantly different amongst all rural sites ($t = 24.239$, d.f. = 5, $p < 0.05$). Amongst urban sites, Tom Campbell's Hill yielded the highest evenness value (0.8814). Amongst rural sites, the maximum value of evenness was found at Sandy Cross: Farm 5A, which also showed the lowest species richness value of rural sites. Generally, comparisons of urban and rural evenness have shown that parasitism among coyotes is similar on a regional scale (urban versus rural), but significantly variable amongst individual sites within urban or rural regions.

Parasite Diversity

Diversity was measured using the Shannon-Weiner index (H'). Expected Shannon-Weiner values of low-to-high species diversity generally range between an H' -value of 1 to 4, respectively (Magurran 2004). We did not log-transform our H' values. We observed no significant difference in species diversity between urban and rural sites ($t = -2.171$, d.f. = 12, $p = 0.051$) (Table 2). The Shannon-Wiener index value for urban areas (1.7726) was similar to rural areas (1.7540). There were significant differences in species diversity amongst rural sites ($t = 36.191$, d.f. = 5, $p < 0.05$), with a maximum Shannon-Wiener value of 1.6829 from Sandy Cross: Farm 5A. Likewise, the diversity of species was significantly different amongst urban sites ($t = 18.402$, d.f. = 7, $p < 0.05$), and Inglewood Wildlands had the maximum Shannon-Wiener value of 1.7618. In sum, the diversity measures of parasitism showed no differences between urban and rural regions of Calgary, while parasite diversity was significantly variable amongst sites within each region.

REGION	SITE	Diversity Indices			t -value			df			p -value		
		Richness (S)	Pielou's Evenness Index (J')	Shannon-Wiener Index (H')	Richness (S)	Evenness (J')	Diversity (H')	Richness (S)	Evenness (J')	Diversity (H')	Richness (S)	Evenness (J')	Diversity (H')
URBAN	ARBOR	5.00	0.8082	1.3008	10.538	30.579	18.402	7	7	7	<0.05	<0.05	<0.05
	NHP	6.00	0.6370	1.1414									
	WEASEL	5.00	0.7843	1.2622									
	STANLEY	5.00	0.7270	1.1700									
	FCPP-W	8.00	0.7425	1.5439									
	FCPP-E	5.00	0.7851	1.2636									
	INGLE	9.00	0.8018	1.7618									
	TCH	5.00	0.8814	1.4185									
RURAL	BHS	10.00	0.6820	1.5703	19.852	24.239	36.191	5	5	5	<0.05	<0.05	<0.05
	HCR	8.00	0.6678	1.3887									
	KERFOOT	9.00	0.7500	1.6478									
	RANCH	9.00	0.6837	1.5022									
	16A	8.00	0.7571	1.5744									
	5A	7.00	0.8648	1.6829									
TOTAL	URBAN	14.00	0.6717	1.7726	-3.308	0.937	-2.171	12	12	12	<0.05	0.367	0.051
	RURAL	15.00	0.6477	1.7540									

Table 2. Measures of diversity of coyote gastrointestinal parasites identified from all urban and rural sites of Calgary, Alberta. Indices were used to compare variation within urban and rural categories, then between categories of urban and rural, $p < 0.05$.

DISCUSSION

Comparisons of Parasite Community to North American Surveys

Few studies of parasitism in coyotes have been conducted in Canada. For those that have been published, parasite species richness or evenness was not explicitly reported. The most recent survey of coyote parasitism in the region was performed by Thompson et al. (2009). Thompson et al. identified *T. leonina*, *Taenia* spp., *Cryptosporidium* spp., and *Giardia* spp. in coyotes. However, the Thompson et al. (2009) study used molecular methods that focused on identifying *Giardia* spp. and *Cryptosporidium* spp., and were not fecal-based; consequently, measures of diversity were not recorded. Another survey of coyote parasitism in Montana by Seese et al. (1983) revealed seven cestodes, nine nematodes, and one trematode species existing in the sampled population. The latter study found that the coyote population in the Montana region showed a parasite richness value of $S = 17$, which is comparable to the $S = 15$ that we documented in the Calgary region.

In Manitoba, Samuel et al. (1978) identified twelve endoparasites in coyotes. Among these parasites, all gastrointestinal nematodes were also found in our study, with the exception of *Oslerus osleri*. Importantly, our flotation methods were limited in the ability to recover *Oslerus* larvae. Gompper et al. (2003) identified nineteen parasites in 145 coyote fecal samples from New York State. In addition, Bridger et al. (2009) identified three taeniid species, *Mesocostoides* spp., *T. canis*, *T. leonina*, *Crenosoma vulpis*, *Physaloptera rara*, *Uncinaria stenocephala*, and *Angiostrongylus vasorum* in Newfoundland. However, these surveys did not explicitly report site-scale richness results, and we were unable to compare variability in site richness to our survey.

Considering evenness, our low values are similar to those observed in other surveys. For example, the dominant prevalence of *T. leonina* in Manitoba (Samuel et al. 1978), Montana (Seese et al. 1983), New York State (Gompper et al. 2003), Alberta (Thompson et al. 2009), and Newfoundland (Bridger et al. 2009) surveys reflected unevenness in reported coyote parasite communities. Because *T. leonina* was also a dominant parasite in our samples, this uneven distribution of parasites in our samples reflects similar values of unevenness to our results, urban or rural.

Parasite Richness

While the component parasite community of coyote fecal samples was similar between urban and rural sites, we found significantly higher parasite richness in rural areas relative to urban areas (Fig. 3). This difference, however, was largely due to the absence of *P. kellicotti* in the urban region, and the parasites present in the entire Calgary region were similar (Neuhauser and Poulin 2004). Yet, richness values for individual rural sites tended to be greater than urban sites. Measures of urban canids in France reflect this result: Robardet et al. (2008) highlighted a decreasing trend of *Echinococcus multilocularis* prevalence in foxes from rural to peri-urban to urban habitats. They attributed the lower urban tapeworm prevalence to the relatively higher consumption of anthropogenic foods rather than small mammals, which may act as intermediate hosts of parasites. While our study measured the entire parasite community – not targeted

parasite species – the life-cycles of the identified gastrointestinal parasites are relatively similar and should be influenced by the same biotic and abiotic factors of exposure.

Diet is a possible influential factor for greater coyote parasite richness in rural rather than urban sites in the Calgary region. For instance, VerCauteren et al. (2008) showed that coyotes were bioaccumulators of *Mycobacterium bovis* through feeding on infected white tailed deer in rural areas. Also, Quinn (1997) identified that voles were the most abundant mammalian food item in the diet of coyotes in a mixed agricultural habitat. The diversity of small mammal species, and other vertebrates, which are potential sources of coyote prey and parasites, is greater in rural habitats. We assessed diet concurrent with our parasite analysis, and a combined analysis will be the subject of another future publication. In Calgary, anthropogenic food sources have been identified as a prominent diet source (Lukasik and Alexander 2012), and could also buffer the richness of parasites in urban versus rural areas. Gastrointestinal parasites are considered macroparasites and have different transmission factors than microparasites such as viruses. Nematodes, for example, can exist in free-living stages and can infect animals that ingest these infective eggs from a fecal-oral route.

Some nematodes also infect intermediate hosts such as small mammals. Coyotes can acquire the parasites by direct contact with contaminated feces that results in ingestion of parasites or indirectly by feeding on infected hosts. Rural areas have a greater diversity of rodent, lagomorph, or other small mammals that can be intermediate hosts of many identified parasites in Calgary. Transmission routes vary depending on the species of parasite, including the species of intermediate host infected, and this may vary in urban environments versus rural environments (Lindenfors et al. 2007). For example, *P. kellicotti* was only documented from the rural Calgary region. *P. kellicotti* is a lung fluke with a complex life cycle associated with two intermediate aquatic invertebrate hosts. An omnivorous or carnivorous mammalian host would be exposed to this parasite by ingesting the second intermediate host, such as a crayfish, in which the infective metacercariae stage develops (Bowman, 2008). The diversity of hosts that exist in rural semi-aquatic habitats is likely greater than urban habitats of Calgary. Consequently, the probability of exposure to this parasite is most likely higher for coyotes in Calgary's surrounding rural area rather than urban environments. Current research on diversity of diet items of urban and rural coyotes in our research group will complement our hypothesis, and help determine whether diet diversity is higher in urban or rural environments and therefore implicated in parasite occurrence.

Some studies that focused on the effect of the urban environment on wildlife parasitism, by comparing urban to rural samples, have found disparate results to ours. For example, it was found that gray foxes were more likely to have canine parvovirus in urban than rural areas of San Francisco (Riley et al. 2004). Similarly, Lehrer et al. (2010) determined that urban woodchucks were more likely to have *Toxoplasma gondii* than rural woodchucks in western Arkansas. Intensive work on the persistence of *E. multilocularis* in Switzerland (Deplazes et al. 2004) also showed this trend in association with urban foxes. This latter study found that factors influencing prevalence of *E. multilocularis* in the urban periphery included: moderate human density (and subsequent anthropogenic food supply), fox density, predation on rodents, environmental *E. multilocularis* egg contamination, recreational and soil-linked activities (farming, gardening), and free-roaming dogs and cats. Yet, in our study, the parasite community was similar in agricultural and recreational areas relative to the central urban area. Because our findings are

inconsistent with that of other regions, the factors that influence coyotes' probability of exposure to parasites might be unique to Calgary. Dietary factors of exposure, specifically, may be a prominent influence of parasite richness in rural than urban areas.

Coyote inter-specific interactions could also facilitate exposure of coyotes to parasites. Fecal-oral transmission is the most direct means for acquisition of macroparasites. Cross-species transmission is a common means for parasite exchange in diverse host communities (Aguirre 2009). Coyotes are definitive hosts for all identified parasites in Calgary, Alberta, with the exception of pinworm. In addition, many other terrestrial species are definitive hosts for the identified parasites (Bowman 2008). Larger mammals existing in rural environments can act as reservoirs for macroparasites. If many host species existing in sylvatic cycles are sharing habitats with coyotes, parasite spillover could take place from sympatric populations. For example, taeniid parasites can exist among a variety of sylvatic cycles from a multitude of intermediate and definitive hosts (Wobeser 2006). Similarly, alveolar hydatid disease, which is caused by *Echinococcus granulosus*, is a parasite maintained by coyotes, wolves, or moose, among other wildlife species: these terrestrial mammals would likely exist at higher densities in rural than urban habitats. In urban areas, domestic dogs and cats are also definitive hosts for some identified parasites and it is possible that parasite spillover could occur between coyotes and domestic canines and felines. This may influence exposure for coyote parasitism in urban sites in Calgary, but the greater parasite richness in rural sites suggests otherwise. Regardless of shared habitats and interactions with sympatric populations in urban and rural areas (Bekoff 2001), all identified parasites are transmitted indirectly and coyotes will most likely be exposed to these parasites by oral ingestion of contaminated feces or through the ingestion of an intermediate host. Thus, diet likely remains a prominent influence on parasite exposure for coyotes. Movement behavior may also increase the probability of exposure between individuals, and these factors are described in the context of evenness and diversity below.

Parasite Evenness and Diversity

Species diversity and evenness, which describe the relative representation of parasite species (identified as eggs/cysts/oocysts) within fecal samples, were not significantly different between urban and rural areas. This suggests that the quantified distribution of parasite prevalence between sites was similar in both areas. In almost all sites, evenness (as expressed by Pielou's Index) was considered low. This is largely a result of the dominant prevalence of *T. leonina*, *Cystoisospora* spp., and *Taenia*-like spp., combined with the lower prevalence of rarer species such as *Diphyllbothrium* spp., *Dipylidium caninum*, *Giardia* spp., *Alaria* spp., or *Paragonimus kellicotti*, which were not identified at each rural or urban site. In terms of parasite prevalence, this reflects an uneven distribution of dominant parasites versus rare parasites in the parasite community (Watve and Sukmar 1995). *T. leonina* and *Cystoisospora* spp., in addition to *Taenia*-like spp. and *Trichuris* spp., were documented in every rural site. For parasite diversity, the Shannon-Wiener index values were less than 2, indicating both urban and rural sites had low diversity. The Shannon-Wiener index consolidates a measure of richness and evenness, and low evenness values (due to dominance by high-prevalence species) can diminish the Shannon-Wiener index values. Dominant representation of high-prevalence parasite species is common for parasite communities of coyotes (Samuel et al. 1978, Lesmeister et al. 2008), and heterogeneity in the distribution of parasites in wild animal populations is not unusual (O'Brien et al. 2009).

The variation in evenness and diversity between individual sites may also be explained by interacting biotic and abiotic factors. In urban environments, inter-specific interactions or contact with contaminated feces are factors that could facilitate direct- or indirect-transmission events in urban areas. For instance, Wright and Gompper (2005) showed that in raccoons, the amount of intra-specific interactions raccoons had strongly influenced the prevalence of gastrointestinal parasites in raccoons. If urban habitats in Calgary provide variability in access and abundance of resources, coyotes could have variability in interaction with other species at these sites, whereby parasite exchange may occur. In addition, abiotic factors such as riparian travel routes, anthropogenic corridors, and use of resource nodes should therefore be further investigated to identify associations of habitat structure with parasite diversity (Randa and Yunger 2006).

Among our chosen urban sites, the Inglewood Wildlands site was the most diverse and species rich ($S = 9$). This site was adjacent to high-density residential land use and exists as a portion of a bird sanctuary along a river meander. The surrounding area is used heavily by domestic animals and humans, and abandoned railroad tracks also enter the Wildlands. The Inglewood Wildlands may be an important foraging area for many species due to the dense urban land use that surrounds it. Because many linear transects and hydrological features lead to this site, the Wildlands may act as a common resource aggregation area, where coyote individuals from different groups may experience greater interaction, both direct or indirect, relative to other urban sampling sites. Coyotes are also known to use river edges or creeks as travel routes (Hilty and Merenlender 2004). In so doing, the species may interact more frequently with domestic pets, people, and other potential hosts (Riley et al. 2004). If coyotes are travelling from urban habitats in close proximity to the Inglewood site then host contact rates may be higher at this site than other urban sites, which may explain the greater richness and diversity of parasite species in the samples we observed in this area (Wright and Gompper 2005, Wobeser 2006). The relatively high prevalence of *T. canis* in Inglewood Wildlands may also shed light on the population structure of the coyote population. For example, *T. canis* eggs are typically shed within animals less than 6 months of age (Bowman 2008). Finally, exposure to this parasite may also be due to the presence of young dogs in the area, though the use of Inglewood Wildlands for domestic canines is prohibited.

As with the Inglewood Wildlands in the urban survey, one rural site exhibited a noticeably higher richness, Big Hill Springs Provincial Park. Occurring in the bottom of a narrow valley, the placement of the access road and creek in a valley bottom may facilitate coyotes to use similar travel paths to many other wildlife species (Hilty and Merenlender 2004). Kunkel and Pletscher (2001) identified that valley bottoms are common, and sometimes preferred, travel routes for wolves in Glacier National Park, Montana. Our variable measures of evenness complement these hypotheses by reflecting the presence and absence of given rare parasite species such as *Alaria* spp. and *Toxocara canis*, and the dominant presence of *Toxascaris leonina* and *Cystoisospora* spp. in urban and rural sites. It would be valuable for future research to examine how landscape structure and connectivity (McCallum 2008) influences parasite parasitism in coyotes in and around Calgary.

CONCLUSIONS

There was variation in parasite communities found in urban and rural environments around the city of Calgary, Alberta. In urban and rural areas of the city, the parasite community carried by coyote hosts was similar, but significant variability in the presence and proportion of different parasites was observed amongst urban and rural sites. If coyotes disperse in and out of urban and rural environments, then transient individuals help maintain parasite infections across broad areas occupied by coyotes. For Calgarians, any attempts to reduce the numbers of coyotes by management efforts would not necessarily result in any significant reduction in the presence of parasites, including high-risk species, if nomadic individuals are moving between metapopulations located at different sites.

Coyotes are an adaptable and resilient species, and may be resistant to factors that promote susceptibility to infection in urban environments, such as stress or poor nutrition. Therefore, though many coyotes are infected with this community of parasites – whether urban or rural – the impacts of these parasites on the status of coyote populations may be low. Some of the more susceptible individuals such as young, senescent, injured, or those that are immune-compromised probably do die as a result of the disease impacts of gastrointestinal parasites (Pence and Windberg 1994). However, it is clear from our data that coyote individuals often act as a reservoir for at least one parasite. Our parasite diversity results clearly show that multiple parasites can be maintained by coyotes, and coyotes with high parasitism may potentially represent healthy individuals, tolerant to infection. Parasite evenness measures indicated that dominant parasites exist among coyote metapopulations, but that rarer parasites exist in specific habitats interspersed among the city and in agricultural habitats as well.

Our research showed variability in parasite species richness, evenness, and diversity. These urban and rural differences have prompted further hypotheses about carnivore movement, dietary behavior, and interspecific interactions between metapopulations. Patterns in urban ecology have been suggested from this method whereby no individual coyotes were directly baited or handled. Also, we chose diversity indices to document our findings in an intuitive and feasible way for the identification of regional-scale patterns. Such diversity measures have shown patterns across individual sites as well as general trends between the urban versus rural regions in Calgary. We suggest that variation in the gastrointestinal parasite community between sites is most strongly attributed to influences of urban landscape structure on biotic and abiotic factors of transmission among coyote groups. In conclusion, our parasite research revealed that urban habitats in Calgary do not predispose coyotes to higher levels of parasitism, and hence the risk of human infection remains low. While few of the parasites we identified have zoonotic potential, further pathogen-targeted methods (i.e., molecular investigations) are required to quantify public health risks. Thus, maintaining and coexisting with the thriving urban coyote population in this region continues to have ecological merit.

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