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Serologic Survey for Diseases in Free-Ranging Coyotes (*Canis latrans*) from Two Ecologically Distinct Areas of Utah

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ABSTRACT: The influence of habitat and associated prey assemblages on the prevalence of canine diseases in covotes (Canis latrans) has received scant attention. From December 1997 through December 1999, we captured 67 coyotes in two ecologically distinct areas of Utah (USA): Deseret Land and Livestock Ranch and US Army Dugway Proving Ground. These areas differ in habitat and prey base. We collected blood samples and tested for evidence of various canine diseases. Prevalence of antibodies against canine parvovirus (CPV) was 100% in the Deseret population and 93% in the Dugway population. All juveniles in both populations had been exposed. We found no difference in the prevalence of antibodies against canine distemper virus (CDV) between the two populations (7% versus 12%; P=0.50). However, we did find an increase in antibodies with age in the Deseret population (P=0.03). Evidence of exposure to canine adenovirus (CAV) was found in both populations (52% and 72%); P=0.08). Prevalence of CAV antibodies was influenced by age on both areas (Deseret: P=0.003; Dugway: P=0.004). Antibodies to Francisella tularensis were low on both areas (2% and 4%). We found a significant difference (P=0.001) in the prevalence of exposure to Yersinia pestis between the two populations: 73% in Deseret compared to 11% in Dugway. This difference is most likely due to the prey species available in the two ecologically distinct study areas.

Key words: Canine adenovirus, canine distemper virus, canine parvovirus, *Canis latrans*, coyote, *Francisella tularensis*, plague, serological survey, tularemia, *Yersinia pestis*.

Surveys for antibodies against viral and bacterial diseases of coyotes (*Canis latrans*) have been conducted in many western states (Thomas et al., 1984; Gese et al., 1997; Cypher et al., 1998; Grinder and

Krausman, 2001). However, comparisons between populations within a state are few. In addition, the influence that differences in habitat types and management practices might have on the prevalence of certain canine diseases has received little attention. Also, the last reported serologic survey of coyotes in Utah (USA) occurred in 1983 (Thomas et al., 1984). Canine parvovirus (CPV) was absent in the free-ranging coyote population until an epizootic in 1979, coinciding with epizootics in domestic dogs (Thomas et al., 1984). By the fall of 1980, CPV was enzootic in Utah. Since then the human population and size of urban centers has increased substantially in the state (30% increase in human population in Utah from 1990 to 2000; US Census Bureau, 2000). A subsequent increase in the domestic dog population has likely occurred in response to this increase in human population. Changes in disease prevalence in the last 20 yr may have occurred with an increase in the dog population and possible greater contact between domestic and wild canids as urban centers expand and wildlife habitat is changed into residential developments in rural areas.

In addition, the US Army Dugway Proving Ground (DPG) was historically used as a facility for testing biological agents such as plague and tularemia. Information on the prevalence of these diseases and other potential canine diseases are necessary documentation for DPG's Environmental Impact Statement. By comparing the prevalence of antibodies against *Yersinia pestis* and *Francisella tularensis* in the coyote population where biological testing occurred, we can determine the long-term impacts biological agent testing has had on the coyote population. We report results of a serologic survey for evidence of antibodies against CPV, canine distemper virus (CDV), canine adenovirus (CAV), *Y. pestis*, and *F. tularensis* in free-ranging coyotes from two ecologically distinct areas in Utah.

We captured coyotes from two ecologically distinct areas in Utah: the US Army Dugway Proving Ground (DPG) and the Deseret Land and Livestock Ranch. Dugway Proving Ground (39°53'-40°24'N, 112°45′–113°43′W) is an isolated US Army installation located 128 km southwest of Salt Lake City and covers 3,330 km² of the Great Basin Desert. Due to its mid-latitude location, this region is often characterized as cold desert. Most of DPG consists of flat terrain with salt playas supporting pickleweed (Allenrolfea occidentalis) and chenopod habitat containing shadscale (Atriplex confertifolia), gray molly (Kochia americana), and greasewood (Sarcobatus vermiculatus). Interspersed among the flat terrain are steep mountain ranges that are cooler, receive more precipitation, and support sagebrush (Artemesia sp.), horsebrush (Tetradymia sp.), and Utah juniper (Juniperus osteosperma) (AGEISS Environmental Inc., 2001). Temperatures range from an average of -8.8 C in winter to 34.7 C in summer. Mean annual precipitation is 20.07 cm. Principle prey items available to covotes include blacktail jackrabbits (Lepus californicus), kangaroo rats (Dipodomys sp.), deer mice (Peromyscus sp.), and cottontail rabbits (Sylvilagus sp.) that reflect the desert environment of DPG (AGEISS Environmental Inc., 2001).

The 400-km² Deseret Land and Livestock Ranch is located in northeastern Utah (41°10'-41°28'N, 111°2'-111°25'W). In contrast to DPG, this study area is primarily sagebrush (*Artemesia tridentate* *wyomingensis*) steppe with an understory of western wheatgrass (Pascopyrum smithii), needle-and-thread grass (Stipa co*mata*), and Indian rice grass (Oryzopsis hymenoides) (Bromley, 2000). Also, unlike DPG, Deseret is located near human populations (Evanston, Wyoming, USA, and Woodruff, Utah) and several large ranches. Average annual rainfall is 27.6 cm. Temperatures range from an average of -9.4C in winter to 15.6 C in summer. Major prey species available to covotes include whitetail jackrabbits (Lepus townsendii), Unita ground squirrels (Spermophilus armatus), and deer mice (Peromyscus maniculatus), plus winter carrion (mainly elk, Cervus elaphus, carcasses) (Bromley, 2000). The main distinction between the two study areas was overall habitat type (cold desert versus sagebrush steppe) and their corresponding difference in the prey community assemblages.

We captured coyotes in the early winter of 1997, 1998, and 1999 at Deseret and in 1999 at DPG using a hand-held net-gun fired from a helicopter (Barrett et al., 1982; Gese et al., 1987). We captured any covote observed on the study area, including entire social groups when possible. Animals were weighed, their sex determined, aged by tooth wear (Gier, 1968), eartagged, and radiocollared (Advanced Telemetry Systems, Inc., Isanti, Minnesota, USA). We extracted a first vestigial premolar from the lower jaw of coyotes captured at Deseret for aging by cementum annuli analysis (Linhart and Knowlton, 1967). We extracted a 10-15 ml blood sample from the cephalic or saphenous vein of captured coyotes. We placed each blood sample into a glass serum tube (Vacutainer, Becton Dickinson, Rutherford, New Jersey, USA) and centrifuged for 30 min. The serum was harvested and stored at -20 C. We classed coyotes as juveniles (<12 mo old) and adults (\geq 12 mo old).

We analyzed serum samples for antibodies against CDV, *Yersinia pestis*, and *Francisella tularensis* at the Wyoming State Veterinary Laboratory (University of Wy-

Study area Age class	n	CPV	CDV	CAV	F. tularensis	Y. pestis
Deseret						
Adult	22	100	23	95	0	86
Juvenile	18	100	0	44	6	56
Dugway						
Adult	21	91	10	67	5	14
Juvenile	6	100	0	0	0	0

TABLE 1. Prevalence of antibodies against canine parvovirus (CPV), canine distemper virus (CDV), canine adenovirus (CAV), *Francisella tularensis*, and *Yersinia pestis* in coyotes on Deseret Land and Livestock (Deseret) and Dugway Proving Ground (Dugway), Utah, 1997–99.

oming, Laramie, Wyoming) and for CPV and CAV antibodies at the Washington Animal Disease Diagnostic Laboratory (Washington State University, Pullman, Washington, USA). Canine distemper virus antibody was detected by the serum virus neutralization test described by Appel and Robson (1973). An antibody titer \geq 1:10 was considered positive for antibodies against CDV. Antibodies against CPV were detected using an indirect fluorescent antibody test (Rose et al., 1992). A titer of >1:25 was considered positive for CPV antibodies. Antibodies against canine adenovirus were detected by the virus neutralization test (Appel et al., 1975). A titer of >1:4 was considered positive. To determine the prevalence of antibodies against Y. pestis, we used passive hemagglutination inhibition (PHI) tests and an enzyme linked immunosorbent assay (ELISA) (Chu, 2000); a titer of >1:16 was considered positive. We used the microscopic agglutination test as described by Gese et al. (1997) for detecting antibodies against F. tularensis; a titer of $\geq 1:127$ was considered positive.

For all statistical tests, we used each individual coyote as the sampling unit. All coyotes were represented by one sample. There were no repeated samples from the same coyote. We used the chi-square (χ^2) test to analyze the prevalence of antibodies among age classes and between sexes within each study area and for all coyotes between the study areas (Zar, 1996). We used a Fisher exact test when the contingency table contained an expected frequency of less than 1.0 in any cell (Zar, 1996). We performed all statistical tests using the computer software program SPSS (SPSS Base 10, Chicago, Illinois, USA).

We collected blood samples from 67 coyotes (41 males and 26 females) from December 1997 to December 1999. We captured 18 juveniles and 22 adults from the Deseret study site. We sampled 18 in 1997, 10 in 1998, and 12 in 1999 at Deseret. In December 1999, we captured 27 coyotes, consisting of six juveniles and 21 adults, at the DPG study site.

We completed laboratory analysis for CPV antibodies on serum samples from all 67 coyotes (Table 1). Coyotes had CPV titers ranging from 1:20 to 1:2,560. Eight juveniles had titers of $\geq 1:1,280$. For all coyotes combined, we found the prevalence of CPV among juveniles (100%, 24/ 24) and adults (95%; 41/43) was not different (χ^2 =1.15, 1 df, P=0.28). On Deseret, all coyotes sampled showed antibodies against CPV (40/40). Canine parvovirus antibodies were also common at DPG, with 93% (25/27) of the population positive. On DPG, there was no significant difference among age classes (Fisher's test, P > 0.60) or between the sexes (Fisher's test, P > 0.70). We found only one juvenile and three adults had evidence of recent exposure (1:1,600). We found no significant difference in prevalence of CPV antibodies between the study areas (Fisher's test, P=0.16), and there was no difference in evidence of recent exposure between the study areas ($\chi^2 = 0.41$, 1 df, P = 0.52).

We completed serology for CDV anti-

bodies on 67 coyotes (Table 1). For all coyotes combined, we found the prevalence of antibodies against CDV was different between juveniles (0%; 0/24) and adults (16%; 7/43) (χ^2 =4.36, 1 df, P=0.037). The prevalence of CDV antibodies increased significantly with age in the Deseret coyotes (χ^2 =4.67, 1 df, *P*=0.031), but was not different between males and females $(\chi^2=0.15, 1 \text{ df}, P=0.70)$. Antibody titers for adults ranged from 1:32 to 1:256. On DPG, we found no juvenile covotes had antibodies against CDV and only two of 19 adults were positive (Fisher's test, P > 0.60). On DPG there was no difference in prevalence of CDV antibodies between the sexes (Fisher's test, P > 0.70). We found no significant difference in antibodies against CDV between Deseret (12%; 5/40)and DPG (7%; 2/27) ($\chi^2=0.45$, 1 df, P = 0.50).

We tested for antibodies against CAV in 67 coyotes (Table 1). We found the prevalence of CAV antibodies was 33% (8/24) among juveniles and 81% (35/43) among adults for all covotes combined ($\chi^2 = 15.48$, 1 df, P=0.0001). Age influenced the prevalence of CAV antibodies in the Deseret $(\chi^2 = 12.92, 1 \text{ df}, P = 0.0003)$ and DPG $(\chi^2 = 8.31, 1 \text{ df}, P = 0.004)$ population. We found no difference in prevalence of antibodies between the sexes at Deseret $(\chi^2 = 1.42, 1 \text{ df}, P = 0.23) \text{ or DPG} (\chi^2 = 0.94,$ 1 df, P=0.33). We found the prevalence of CAV antibodies was 72% (29/40) and 52% (14/27) on the Deseret and DPG study areas, respectively ($\chi^2 = 2.99$, 1 df, P = 0.08).

We analyzed 67 coyote serum samples for antibodies against *F. tularensis*. We found only one pup in the Deseret population (1:256) and one adult from the DPG population (1:128) had antibodies for *F. tularensis*. For all coyotes combined, there was no difference in the prevalence of antibodies against *F. tularensis* between juvenile (4%; 1/24) and adult coyotes (2%; 1/43) (χ^2 =0.18, 1 df, *P*=0.67). Comparing between the two study areas, the prevalence of antibodies for *F. tularensis* was 2% (1/40) in Deseret and 4% (1/27) in DPG (χ^2 =0.08, 1 df, P=0.77).

We analyzed serum samples from all 67 covotes for antibodies against Y. pestis. However, we could not determine exact PHI titers for seven serum samples from Deseret, but antibodies were detected by ELISA. For all covotes, we found the prevalence of antibodies against Y. pestis was 42% (10/24) and 51% (22/43) for juvenile and adult covotes, respectively $(\chi^2 = 0.55, 1 \text{ df}, P = 0.09)$. Prevalence of antibody titers to Y. pestis increased with age in the Deseret population ($\chi^2 = 4.71$, 1 df, P=0.03), but was not different between the sexes ($\chi^2 = 1.42$, 1 df, P = 0.23). In contrast to the Deseret area, antibodies for Y. *pestis* were not as prevalent in the DPG population with three of 21 adults positive and no juveniles positive (Fisher's test, P > 0.45). There was no difference between the sexes ($\chi^2 = 0.30$, 1 df, P = 0.59). Positive titers for the adults ranged from 1:32 to 1: 128. We found a significant difference between Deseret (72%; 29/40) and DPG (11%; 3/27) in the prevalence of Y. pestis antibodies ($\chi^2 = 24.35$, 1 df, P = 0.0001).

Serologic evidence of exposure to canine parvovirus was first detected in Utah in 1979, at which time it was found in >70% of a wild coyote population (Thomas et al., 1984). Canine parvovirus is well established in both Utah populations (93% DPG and 100% Deseret), and is among the highest reported (Thomas et al., 1984; Cypher et al., 1998; Gese et al., 1991; Holzman et al., 1992). All juveniles in both populations had antibodies to CPV which indicated the virus was being transmitted among the covotes in multiple years and thus could be considered endemic. These results are similar to the northern Yellowstone population where exposure was 100% in all coyotes, except pups <3months old (Gese et al., 1997). High prevalence of antibodies is often associated with a highly contagious but non-fatal infection because prevalence is measured among survivors (Thomas et al., 1984). We did not capture juveniles until they were >7 months old (after the majority of pup mortality due to CPV would have occurred), and therefore, do not know to what extent CPV may have on limiting recruitment into the two populations.

Canine distemper virus antibody prevalence was low in both the Deseret (12%)and DPG covote population (7%), and antibody titers were low. These results suggest that CDV had not been active in these populations in the past few years. Our findings of prevalence of antibodies to CDV are among the lowest reported in free-ranging coyotes (Trainer and Knowlton, 1968; Guo et al., 1986; Gese et al., 1991, 1997; Cypher et al., 1998). The prevalence was not different between the sexes, but did increase with age in both Utah populations (i.e., no pups were positive). Canine distemper virus may cause mortality in young pups (Gier and Ameel, 1959; Gier et al., 1978), but some probably survive.

Canine adenovirus-1 (infectious canine hepatitis) antibody prevalence increased with age in both populations, similar to findings by Gese et al. (1997) and Cypher et al. (1998). Prevalence of CAV in the Utah populations is similar to prevalence in other coyote populations (Trainer and Knowlton, 1968; Holzman et al., 1992; Cypher et al., 1998). Both CDV and CAV antibody prevalence increased with age, whereas CPV antibodies were found in all age classes. One possible effect of these canine diseases is reduced pup survival. However, others have suggested that these diseases exist in an enzootic state within covote populations (Thomas et al., 1984; Guo et al., 1986) and may only cause significant mortality during stressful conditions such as food scarcity, high density, or parasitism (Trainer and Knowlton, 1968).

The most interesting difference between the two populations was the evidence of antibodies against Y. *pestis* in the Deseret population (73%) as compared to the DPG population (11%). High antibody prevalence of Y. *pestis* has been found in other studies (Barnes, 1982; Gese et al., 1997). Coyotes rarely serve as a reservoir for transmission of plague to other species (Von Reyn et al., 1976; Barnes, 1982). Coyotes do not usually develop clinical signs when infected (Von Reyn et al., 1976). However, coyotes do develop antibodies that can last up to 6 mo or more making them an excellent sentinel species for plague (Barnes, 1982). Serologic testing of these carnivores can help establish the presence of plague among local rodent populations (Willeberg et al., 1979; Thomas and Hughes, 1992).

Habitat and its influence on the prey community was apparently associated with the difference in the serum antibody prevalence of Y. pestis in the two Utah populations. Ground squirrels are relatively abundant on the Deseret study area (Bromley, 2000). Prairie dogs (Cynomys sp.) and ground squirrels are often affected by plague in western US. In contrast to Deseret, ground squirrel populations on DPG are extremely low and have only recently begun to show an increase in population size (AGEISS Environmental Inc., 2001). The cold desert environment and chenopod habitat characteristic of most of the DPG is not optimal habitat for ground squirrels. Messick et al. (1983) noted that Peromyscus and Dipodomys have been suspected as being plague reservoirs in Utah. Although, these two species are common on DPG and in the covote diet (Kozlowski, unpubl. data), the coyotes of DPG have low prevalence to Y. pestis. Plague was endemic to the extreme western portion of DPG in 1952, but was usually found above 1,829 m (Stark, 1958). The Deseret study area contains the preferred habitat for ground squirrels and therefore may have more plague present in the prey community which is reflected in the prevalence of Y. pestis in the coyote population.

Dugway Proving Ground was historically used as a testing facility for biological agents including tularemia and plague. Serologic testing of several species, including coyotes was conducted during periods of open-air testing. However, only one species, an Ord's kangaroo rat (*D. ordii*), had antibodies against *Y. pestis*, while 13 jackrabbits (*L. californicus*) and one coyote had antibodies against *F. tularensis* (Vest et al., 1965). Sampling occurred in a wide area surrounding and on DPG, so it is not clear if these animals were on DPG, or in areas surrounding the military base. Based on current serologic results, exposure to *F. tularensis* appears to be uncommon.

Intrastate comparisons of diseases among coyotes are few. Also, coyotes in Utah have not been sampled for disease prevalence since 1983 (Thomas et al., 1984). Periodic sampling for diseases among carnivores may be beneficial to wildlife managers and biologists by documenting changes in disease prevalence that occurs with habitat differences, landscape changes, and human encroachment into wildlife habitat as urban centers expand.

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