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## SEROLOGIC SURVEY AND SERUM BIOCHEMICAL REFERENCE RANGES OF THE FREE-RANGING MOUNTAIN LION (*FELIS CONCOLOR*) IN CALIFORNIA

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**ABSTRACT:** Serum samples from 58 mountain lions (*Felis concolor*) in California (USA) were collected between April 1987 and February 1990. Nineteen serum samples were used for serum biochemistry determinations; the ranges were similar to reference values in domestic cats, captive exotic felidae and free-ranging mountain lions. A serological survey was conducted to determine whether antibodies were present against selected infectious agents. Fifty-four (93%) of 58 sera had antibodies against feline panleukopenia virus. Fifteen (68%) of 22, 16 (28%) of 58, 11 (19%) of 58, and 10 (17%) of 58 had serum antibodies against feline reovirus, feline coronavirus, feline herpes virus, and feline calicivirus, respectively. Twenty-three (40%) of 58 and 21 (58%) of 36 had serum antibodies against *Yersinia pestis* and *Toxoplasma gondii*, respectively. Only one of 22 sera had antibodies against the somatic antigen of *Dirofilaria immitis*. Feline leukemia virus and feline immunodeficiency virus antigens were not detected in any mountain lion's sera. All 58 sera samples were negative for antibodies to feline immunodeficiency virus and *Chlamydia psittaci*.

**Key words:** Mountain lion, *Felis concolor*, serum biochemistry, serological survey, feline panleukopenia virus, reovirus, coronavirus, herpes virus, calicivirus, feline leukemia virus, feline immunodeficiency virus.

### INTRODUCTION

Mountain lions (*Felis concolor*) are widely distributed in rural, agricultural, and rangeland areas of California (USA), occupying approximately 207,000 km<sup>2</sup> of habitat (Mansfield and Weaver, 1989) in all three general climatic zones within the state (Riemann et al., 1978). It is the largest free-ranging felid in California. The population in California is estimated to be  $\geq 5,100$  animals (Mansfield and Weaver, 1989). The impact of the mountain lion on wild prey species and California's livestock has led to numerous management regulations affecting mountain lion populations (Weaver, 1982; Mansfield and Weaver, 1989). Gaining information about exposure of the mountain lion to potential infectious agents may assist in future studies regarding the impact of these diseases on the population and provide baseline information for future management programs. Our objective was to determine the

presence of antibodies against 11 infectious agents that occur in domestic cats and wild carnivores in California. Serum biochemical values were determined to establish a reference range for the free-ranging population in California.

### MATERIALS AND METHODS

Whole blood for serum samples from 58 mountain lions was collected by personnel of the California Department of Fish and Game (Sacramento, California) between April 1987 and February 1990. Samples were collected from all three general climatic zones in California (Riemann, 1978): the coastal range ( $n = 12$ ), the central valley ( $n = 29$ ), and mountain high desert ( $n = 17$ ). The distribution of samples collected by county was San Diego County (33°40'N, 117°40'W),  $n = 6$ ; Los Angeles County (34°40'N, 118°50'W),  $n = 1$ ; Orange County (33°60'N, 117°70'W),  $n = 5$ ; Fresno County (37°10'N, 119°20'W),  $n = 12$ ; Stanislaus County (37°60'N, 121°00'W),  $n = 3$ ; Sacramento County (38°50'N, 121°30'W),  $n = 1$ ; San Joaquin County (38°00'N, 121°30'W),  $n = 1$ ; Madera County (37°50'N, 119°70'W),  $n = 1$ ; Kern County (34°90'N,

118°80'W),  $n = 11$ ; El Dorado County (38°70'N, 120°50'W),  $n = 4$ ; Sierra County (39°50'N, 120°20'W),  $n = 6$ ; Placer County (39°00'N, 120°70'W),  $n = 3$ ; Mono County (37°90'N, 119°10'W),  $n = 1$ ; and Riverside County (33°60'N, 116°70'W),  $n = 3$ . The sample population consisted of 34 females, 22 males and two of unrecorded gender. Age distribution included 44 adults ( $\geq 2$ -yr-old), 10 animals  $\leq 1.5$ -yr-old, and four unrecorded ages. Professional trackers used hounds to tree the lions in most captures; leg snares also were used. Animals were being captured as part of several different studies assessing mountain lion populations or for removal of urban nuisance animals. Subsequently they were immobilized with a 5:1 combination of ketamine (Ketaset, Bristol Laboratories, Syracuse, New York, USA) and xylazine (Rompun, Haver-Lockhart Laboratories, Shawnee, Kansas, USA) (Jessup, 1983) at approximately 7.26 to 7.7 mg/kg ketamine and 0.88 to 0.99 mg/kg xylazine for physical examination, sampling, and for those animals that were radio-collared. No abnormalities were noted during physical examinations of the immobilized adults or subadults except for one animal with a corneal ulcer. One kitten (estimated to be 8 to 10 wk of age) was captured with signs of a neurological disorder. Additional diagnostic tests were done by the Zoological Medicine Service, Veterinary Medical Teaching Hospital (VMTH) (University of California, Davis, California, USA). The cerebrospinal fluid (CSF) analysis evaluated appearance, refractive index, red blood cells (RBC/ $\mu$ l), nucleated cells/ $\mu$ l, protein (mg/dl), and a cytology smear (Brobst and Bryan, 1989). The cells for cytology were collected using a cytocentrifuge (Cytospin-2, Shandon Southern Instruments, Inc., Sewickley, Pennsylvania, USA). The myelographic study included plain radiographs plus radiographs following the injection of 2.3 cc iopamidol (41%) (Isovue 200, Squibb Diagnostics, Princeton, New Jersey, USA) into the subarachnoid space at the level of lumbar vertebrae 6–7 (Widmer and Blevins, 1991).

A 30 cc blood sample was drawn from a peripheral vein and placed into sterile serum clot tubes. Blood samples were placed on ice when possible and then centrifuged within 24 hr for serum extraction. Sera were aliquoted into plastic screw top vials and stored at  $-20^{\circ}\text{C}$  until analyzed.

Twenty-one serum samples, collected between April 1987 and April 1988, were used for serum biochemistry determinations. Thirteen parameters (Table 1) were analyzed for each sample by the Clinical Biochemistry Laboratory, Veterinary Medical Teaching Hospital (VMTH) (University of California, Davis, Cal-

ifornia, USA). Serum analysis was done by a Dacos analyzer (Coulter Instruments, Hialeah, Florida, USA). Two samples were excluded from statistical calculations due to extreme variation from expected values. Nineteen values were used to calculate the reference ranges. The mean and standard deviation (SD) were calculated for each parameter. Any value that differed from the mean by greater than 3 SD was classified as an "outlier" and excluded from further calculations (Werner and Marsh, 1975). The mean and SD for each parameter were then recalculated after removal of the outliers. This statistical method was previously reported by Seal et al. (1975) and in the International Species Inventory System, Physiological Data Summary (Anonymous, 1982).

A variable number of sera were submitted for serological testing for each test (Table 2). Twenty-two samples collected between 2 April 1987 and 1 May 1988 were analyzed by 9 May 1988; 36 samples collected from 16 May 1988 through 2 February 1990 were analyzed by 15 February 1990. The same laboratory and assay methods were used whenever possible.

Serum antibodies against feline panleukopenia virus (FPLV) were detected by virus neutralization (VN) tests at either the National Veterinary Services Laboratory (NVSL), Ames, Iowa (USA) using the ICK33 strain of feline parvovirus or the Immunology/Virology Diagnostic Laboratory (VMTH, University of California, Davis, California) using the Philip Roxane strain (King and Croghan, 1965). Antibody titers of  $>1:10$  from NVSL or  $1:8$  from University of California, Davis (UCD) were considered positive.

Virus neutralization antibodies against feline reovirus (FRV) using a canine isolate were examined at the NVSL, Ames, Iowa. Antibody titers  $>1:5$  were regarded as positive.

Testing for antibodies against feline herpes virus (FHV), feline calicivirus (FCV), feline coronavirus (CV), feline immunodeficiency virus (FIV) and feline leukemia virus (FeLV) were conducted by the Immunology/Virology Diagnostic Laboratory (VMTH, UCD, Davis, California, USA).

Standard VN tests were used to detect antibodies against FHV and FCV (Cottral, 1978). Briefly, for FHV, (100 TCID<sub>50</sub>) herpes strain C-27 were mixed with various dilutions of test serum and incubated on Crandell feline kidney (CrFK) cells (American Type culture collection, Rockville, Maryland, USA) (Crandell, 1958). Titers were considered positive at  $\geq 1:4$ . The calicivirus assay was similar, but used CFI/68FIV strain of picorna virus (Crandell, 1958). Titers were determined positive at  $\geq 1:4$ .

Antibodies to CV were detected by indirect

TABLE 1. Mean values and reference ranges for serum biochemistry of free-ranging mountain lions ( $n = 19$ ) in California, 1987 to 1990.

Parameter	Units	Mean $\pm$ 2 standard deviations	Range
CO <sub>2</sub>	mmol/liter	12.53 $\pm$ 3.49	7–18
Calcium	mg/dl	9.53 $\pm$ 1.72	6.3–11.9
Phosphorous	mg/dl	5.66 $\pm$ 2.30	2.9–12.0
Creatine	mg/dl	2.05 $\pm$ 0.91	0.7–4.0
Blood urea nitrogen	mg/dl	32.89 $\pm$ 12.83	16–58
Glucose	mg/dl	110.58 $\pm$ 74.63	28–309
Alanine aminotransferase	IU/liter	58.78 $\pm$ 33.41	10–138
Alkaline phosphatase	IU/liter	22.63 $\pm$ 22.59	2–82
Total protein	g/dl	6.58 $\pm$ 1.35	4.4–9.1
Albumin	g/dl	3.13 $\pm$ 0.65	1.9–4.6
Globulin	g/dl	3.45–0.83	2.5–5.0
Total bilirubin	mg/dl	0.30–0.50	0.0–1.0
Cholesterol	mg/dl	155.05–59.83	38–275
A:G ratio	—	0.93–0.18	0.68–1.28
U:C ratio	—	20.03–11.58	8.6–41.4

fluorescent antibody (IFA) (Cottral, 1978) utilizing CrFK cells infected with the DF2 strain of coronavirus. Titers were reported as positive at dilutions of  $\geq 1:25$ .

All sera were tested for both FIV antibody and FIV antigen. Antibodies to FIV were detected by IFA using CrFK cells infected with the Petaluma strain of FIV (Yamamoto et al., 1989). The FIV antigen detection utilized a monoclonal p28 core antigen applied in an ELISA (Yamamoto et al., 1989). All sera were tested for viral core proteins FeLV-p27 using a commercially available ELISA kit (DiaSystems–

FeLV: Tech America Diagnostics, Elwood, Kansas; or Synbiotics Corp., San Diego, California).

Twenty-two sera were tested for antibodies to *Chlamydia psittaci* at NVSL, Ames, Iowa. The test was a modified complement fixation assay (Boulanger et al., 1967) utilizing a parakeet strain 6BC at a 1:10 dilution. Thirty-six sera were assayed for *C. psittaci* antibodies at Texas A&M Veterinary Diagnostic Laboratory (Texas A&M University, College Station, Texas, USA) using IFA with cell cultures infected with chlamydia of turkey origin (Kawamura, 1969).

TABLE 2. Results of the mountain lion (*Felis concolor*) serological survey, California, 1987 to 1990.

Infectious <sup>a</sup> agent	Test <sup>b</sup>	Number tested	Number positive	Percent positive	Titer range
FPLV	VN: Strain ICK33	22	22	100	1:11–>1:625
	VN: Philip Roxane Strain	36	32	89	1:8– $\geq$ 1:256
FRV	VN: Canine Strain	22	15	68	1:11–1:280
CV	IFA: Strain FIP DF2	58	16	28	1:25–1:100
FHV	VN: Strain c27	58	11	19	1:4–1:128
FCV	VN: Strain CF1/68FIV	58	10	17	1:4–1:64
FeLV	ELISA: Viral core FeLV p27	58	0	0	—
FIV	IFA: Petaluma Strain	58	0	0	—
	ELISA: Viral core FIV p28	58	0	0	—
<i>Yersinia pestis</i>	HA	58	23	40	1:128–1:4,096
<i>Chlamydia psittaci</i>	CF	22	0	0	—
	IFA	34	0	0	—
<i>Toxoplasma gondii</i>	Indirect Latex Agglutination	36	21	58	1:32–1:4,096
<i>Dirofilaria immitis</i>	IFA: Cuticular/Somatic	22	0/1	0/4	1:40

<sup>a</sup> FPLV, feline panleukopenia virus; FRV, feline reovirus; CV, coronavirus; FHV, feline herpesvirus; FCV, feline calicivirus; FeLV, feline leukemia virus; FIV, feline immunodeficiency virus.

<sup>b</sup> VN, viral neutralization; IFA, indirect fluorescent antibody; ELISA, enzyme-linked immunosorbent assay; HA, hemagglutination; CF, complement fixation.

Passive hemagglutination tests (HA) for antibodies against *Yersinia pestis* were conducted by the Plague Branch (Centers for Disease Control, Fort Collins, Colorado, USA) using procedures recommended by the World Health Organization (Bahmanyar and Cavanaugh, 1976). Titers of  $\geq 1:16$  were considered positive.

Thirty-six serum samples were analyzed for *Toxoplasma* agglutinating antibodies using a commercial indirect latex agglutination test (Kobayashi et al., 1977) Toxotest-MT (Syn-Kit, Inc., Chatsworth, California) at the Department of Veterinary Epidemiology and Preventive Medicine (University of California, Davis, California). Titers of  $\geq 1:32$  were considered positive.

The Department of Veterinary Microbiology (School of Veterinary Medicine, University of California, Davis) used an IFA test for 22 samples to identify cuticular and somatic antigens of *Dirofilaria immitis* (Wong and Suter, 1979). Results were reported for each component at dilutions of 1:20 and 1:40.

Antibody prevalence of each antigen was initially assessed by pooling all years. This was then stratified by season, sex and location. Seasons were defined as winter, 22 December to 21 March; summer, 22 March to 21 June; spring, 22 June to 21 September; and fall, 22 September to 21 December. Location was divided into three general geographic and climatic zones within California; the coastal range, central valley and the mountain/high desert region. Statistical analyses to determine differences in overall antibody prevalence between regions and seasons were done using the Kruskal-Wallis one way nonparametric analysis of variance (ANOVA) (Daniel, 1987).

## RESULTS

Several biochemistry parameters had wide ranges (Table 1).

No significant differences were found in the antibody prevalence to each antigen in mountain lions in California when analyzed by geographic region, season, or sex. Serum antibody titers for FPLV tested at NVSL all were positive and ranged from 1:11 to  $>1:625$ . The one sample  $>1:625$  was from a 8 to 10-wk-old kitten captured because of apparent neurological impairment. Of the 36 sera tested at UCD, 30 (86%) had titers  $\geq 1:256$ . Antibody titers to FRV were positive in 15 (68%) of 22 serum samples collected in 1987 and 1988.

Values ranged from 1:11 to 1:280. Antibodies to FRV were not evaluated after 1988. Titers to CV ranged from 1:25 to 1:100 in 16 of the 58 sera tested. Ten samples collected in 1987 were positive, the remaining six positive samples were collected in early 1988 and all samples collected from June 1988 through February 1990 were negative.

Eleven (19%) of 58 serum samples were positive to FHV at dilutions ranging from 1:4 to 1:128. Forty sera were negative at 1:4 and seven sera could only be read as negative at dilutions of 1:32 or less due to toxic changes in the cell cultures. All sera collected from 1 May 1988 through 2 February 1990 and tested in the second batch of samples were negative for FHV.

Ten (17%) of 58 animals had FCV titers ranging from 1:4 to 1:64. Forty sera were negative at 1:4 and eight sera could only be read as negative at 1:32 or less due to toxic changes in cell cultures. All sera collected prior to April 1988 were negative for FCV. All serum samples were negative for FeLV viral core antigen and negative for FIV viral group-specific antigen (p28) and antibody.

Hemagglutination tests for antibodies against *Yersinia pestis* all were negative until June 1988 (23 samples). From 17 June 1988 through 2 February 1990, 23 (66%) of 35 samples were positive. Fifteen titers were  $\geq 1:128$ , with the four highest samples ranging 1:1,024 to 1:4,096. All sera tested for antibodies against *C. psittaci* using IFA or CF were negative.

The prevalence for *Toxoplasma* serum IgG using a microagglutination assay was 58% (21 of 36). Ten of the 21 positive titers ranged from 1:128 to 1:1,024. Two samples had titers greater than the limit of the test kit, 1:4,096.

Twenty-two samples from April 1987 to May 1988 were tested for *Dirofilaria immitis*. All sera were negative for cuticular antigen and one serum was positive at 1:40 for somatic antigen of *D. immitis*. Whole blood from this animal was tested for circulating microfilariae and none were found.

## DISCUSSION

Some blood parameters of free-ranging mountain lions have been previously reported to differ from captive populations (Currier and Russell, 1982). These differences have been attributed to nutritional factors as well as the exertion and immobilization associated with sampling wild animals. Reference mean values in this report are similar to reference values in domestic cats (Tasker, 1978), captive exotic felidae (Wallach and Boever, 1983; Fowler, 1986), and other reports of captive and free-ranging mountain lions (Currier and Russell, 1982). Several parameters had wide ranges of values which may reflect variation in handling of samples at field sites. The alkaline phosphatase values  $>50$  u/L were collected from animals  $\leq 2$  yr of age. The wide range of glucose values in this study may reflect variation in the length of pursuit prior to capture as well as variable time periods between blood collection and serum separation.

Feline panleukopenia virus was the most prevalent antibody detected in the sample population of mountain lions in California; this is a highly contagious virus affecting most members of the family Felidae, including *F. concolor* (Torres, 1941). Feline panleukopenia virus has been isolated in captive leopards (*Panthera pardus*) (Johnson, 1964), lions (*Panthera leo*) (Studdert et al., 1973), and tigers (*Panthera tigris*) (Povey and Davis, 1977) as a naturally occurring disease. The FPLV antibody titers also have been reported in exotic cats with no evidence of clinical disease or vaccination history (Bush et al., 1981). Cross reactions of the antibody to various parvoviruses such as mink enteritis virus, canine parvovirus, and raccoon parvovirus can occur. Therefore, serum neutralization titers do not distinguish the feline virus. Based on the high prevalence of positive FPLV titers, we suggest an endemic exposure to a parvo-like virus common to free-ranging mountain lions in California. Feline panleukopenia virus is one of the

few feline viruses that persists outside the host, thus allowing animals to be infected by fomites (Scott, 1987). The FPLV is resistant to most environmental temperatures, freezing and drying, and can survive for several years under adverse conditions (Scott, 1987). Virus neutralization titers  $>1:8$  provide protection in domestic cats (King and Croghan, 1965). In domestic cats, clinical signs of FPLV are most severe in animal  $<6$  mo of age (Pedersen, 1988). Adults generally manifest a transient fever and depression. The FPLV affecting late term fetuses in utero can result in cerebellar hypoplasia in the neonate (Pedersen, 1988).

During this study, a mountain lion kitten was documented to have neurological deficits: ataxia, posterior paresis, mild hypermetria, and intention tremors of the head. The single FPLV titer was extremely high (greater than 1:625). The CSF analysis was within normal limits for a domestic cat (Brobst and Bryan, 1989). The myelographic study was unremarkable. Necropsy results were nonspecific and did not contradict the presumptive diagnosis of perinatal FPLV infection.

The 68% antibody prevalence to FRV in this study was similar to that reported by Scott et al. (1970) wherein low FRV antibody titers were detected in 50% of 110 rural domestic cats in New York. Infected domestic cats produced neutralizing antibody titers ranging from 1:30 to 1:60 (Scott et al., 1970) and 13 of the 15 positive mountain lion titers were 1:56 or greater. Feline reovirus has a worldwide distribution and like other reoviruses, causes disease only under special circumstances.

Feline enteric coronavirus (FECV) and feline infectious peritonitis virus (FIPV) are morphologically and antigenically similar and may be different biotypes of a single prototypical virus (Evermann et al., 1991). Current serological tests cannot differentiate between exposure of a cat to FIPV or FECV, therefore the general term CV is used to indicate viral antibody to

either virus. The FECV causes mild intestinal infections and has worldwide distribution in domestic cat populations. Many antibody-positive domestic cats shed FECV, conversely FIPV usually is a fatal disease of domestic and wild Felidae (Pedersen, 1987; Fowler, 1986).

Clinical feline infectious peritonitis (FIP) has been recognized in several exotic felid species (Worley, 1987). Isolation of coronavirus from exotic felids has been reported in cheetahs that died from FIP or a coronavirus-associated disease (Evermann et al., 1988). The high mortality rate associated with FIP makes seropositive survivors rare although antibody conversion without clinical disease after exposure to low virulence FIPV strains has occurred experimentally (Pedersen, 1985). Antibody production in the absence of clinical disease may reflect exposure to FECV or a less virulent FIPV. There has not been any documented isolation of FECV from an exotic felid (Worley, 1987).

Twenty-eight percent of the study population had antibodies to CV. It is unlikely that these antibodies represent exposure to FIPV due to the high mortality associated with this strain. They may represent infection with a nonpathological but antigenically related coronavirus. It is postulated that CV antibody persistence indicates viral persistence (Pedersen, 1987); therefore, the seronegative population of mountain lions probably were CV-free at the time of serum collection.

Feline herpes virus has a worldwide distribution and diagnosis of FHV in several species of captive exotic felids has been based on clinical signs (Bartz and Montali, 1987). Herpes virus has been isolated from captive clouded leopards (*Felis nebulosa*) (Boever et al., 1977), lions (Truyen et al., 1990), and cheetahs (*Acinonyx jubatus*) (Thompson and Sabine, 1971). Feline herpesvirus in domestic cats characteristically elicits modest titers of neutralizing antibody (1:4 to 1:32) and titers decline to minimal levels within 2 mo (Hoover, 1987). Most of the mountain lion titers were at

this low level suggesting past exposure and recovery.

Domestic cats with persistently high FHV titers may be carriers of latent viral infection and subject to periodic reactivation of viral replication (Gaskell and Povey, 1982). One mountain lion may represent a similar form of carrier because the titer was relatively high (1:128) and was unassociated with clinical signs at the time of immobilization. Carrier cats or cats with clinically active infection are the principle sources of virus; infection requires close contact between shedding and susceptible cats (Pedersen, 1988). In domestic cats, virus activation occurs from parturition and lactation and this may be an important source of infection for kittens (Gaskell and Povey, 1982). The solitary habits of the mountain lion may explain the low antibody prevalence to FHV. Alternatively, low antibody prevalence to a domestic cat antigen is difficult to interpret in an untested species such as the mountain lion.

Feline calicivirus has a worldwide distribution in domestic cats and has a tendency to persist as an active asymptomatic infection in up to one-third of adult domestic cats (Pedersen, 1987). The natural fragility of FCV makes persistent carrier-state infections mandatory for the survival of the virus (Schaffer et al., 1980). Direct contact with oropharyngeal secretions is probably the primary mode of transmission (Bartz and Montali, 1987). Isolation of the virus from exotic felids has been successful only in captive cheetahs (Sabine and Hyne, 1970; Simmons, 1971). Based on the low antibody prevalence (17%), we suggest a low level of exposure to FCV or a low level of cross-reactivity with an antigenically related virus.

In this survey, all mountain lion sera were negative for FeLV viral core protein; thus FeLV probably is not enzootic in the population. Reports of FeLV infection in exotic felids are rare and predominantly in captive animals (Briggs and Ott, 1986; Citino, 1986). There is a recent report of

FeLV infection in a free-ranging mountain lion in California which occurred after the present study was conducted (Jessup et al., 1993). False positive FeLV antigen results have been reported in the Florida panther (*Felis concolor coryi*) following administration of a mouse brain-origin, killed virus rabies vaccine (Lopez, 1988).

Feline immunodeficiency virus antibodies have been reported in several species of non-domestic felids, including six (30%) of 20 tested free-ranging Florida panthers captured during 1986 to 1988 (Barr et al., 1989). Detection of FIV antibody is highly correlated with FIV infection (Yamamoto et al., 1989). Several methods are available for detecting FIV antibody and only the IFA was used in this report. When FIV antibodies were demonstrated in the Florida panther, three different assays were used, including IFA, and all three gave the same results (Barr et al., 1989). The negative IFA results in the present study are supported by the negative ELISA results of all 58 samples for FIV antigen (p28) which is a highly conserved antigen. The lack of antibodies in the California population may reflect lack of exposure to these viruses or lack of immunological response if the virus were contacted. Recently, Olmstead et al. (1992) reported nine of 16 free-ranging mountain lions in California had antibodies against FIV using an immunoblot technique. Because the source and method of collection of these samples was never stated, the epidemiological implications of these findings are uncertain.

*Yersinia pestis* has fluctuating activity in wildlife populations. Plague can remain latent in reservoir hosts and recrudesce with changes in climate or rodent populations. Domestic cats (and presumably free-ranging mountain lions) become infected with *Y. pestis* from the bite of infected fleas or from ingestion of infected rodents (Rosser, 1987). Carnivores are valuable sentinels in the detection of *Y. pestis* in wildlife populations (Willeberg et al., 1979). High ti-

ters indicate recent or heavy exposure. This mountain lion survey indicates a change in *Y. pestis* exposure occurred during 1988, when high positive titers were first detected. Positive titers were recorded for all samples collected between May 1988 and February 1990.

Plague in domestic cats usually is an acute, fatal disease characterized by lymphadenopathy and bacteremia. It is unusual for a domestic cat to recover from infection with *Y. pestis* (Raffo, 1986). Titers in domestic cats that survive remain detectable for  $\leq 300$  days (Rust et al., 1971) and may be as long-lived in other felids. Based on the high antibody prevalence to *Y. pestis* in the surveyed mountain lion population, we suggest that some survive exposure, especially since all animals examined were clinically normal. *Yersinia pestis* has been cultured from the brain of a mountain lion from San Diego County, California (not included in this study group) displaying abnormal behavior (D. A. Jessup, pers. comm.).

*Chlamydia psittaci* is a widespread upper-respiratory disease in domestic cat populations. The CF and IFA, using two different sets of sera, both were negative for antibody. Low seroprevalence is not unexpected for *C. psittaci* since immunity generally is weak and of short duration (Hoover, 1987; Pedersen, 1988).

A high antibody prevalence (60%) to *Toxoplasma gondii* was expected in free-ranging felids since the parasite's sexual cycle is dependent on Felidae (Dubey, 1986). Of the 21 positive mountain lions, 18 had titers of  $\geq 1:64$  with five of those titers  $\geq 1:1,024$ . The antibody prevalence to toxoplasma found in this study was in agreement with a study by Riemann et al. (1978) that showed 37% prevalence in wild carnivores in California. The widespread distribution of positive animals also supports the conclusion that the organism is capable of cycling through all climatic regions of the state (Riemann et al., 1978).

Clinical disease associated with *T. gondii* usually occurs in young domestic cats,



is relatively rare, and can cause diarrhea, pneumonia, retinal damage, and abortion (Frenkel et al., 1987). Toxoplasmosis or antibodies to *T. gondii* have been found in Pallas cats (*Felis manul*) (Riemann et al., 1974), bobcats (*Lynx rufus*) (Riemann et al., 1975), mountain lion (Riemann et al., 1978), ocelot (*Felis pardalis*), jaguarundi (*Felis yaguaroundi*) (Jewell et al., 1972), and other wild felids (Dreesen, 1990). It is likely that most free-ranging and feral felidae have antibodies to this organism. The mountain lion may serve as a source of contamination, but like domestic cats, shedding of oocysts probably is minimal after developing an immune response (Frenkel et al., 1987).

The dog is a definitive host and main reservoir for *Dirofilaria immitis*. Domestic cats are a susceptible but resistant host for the filariid worm (Pedersen, 1988). Cats generate a vigorous immune response to all stages of the parasite, especially migrating pre-adult forms. The immune response is directed mainly at the cuticular proteins of the parasite and a positive IFA against cuticular antigens is diagnostic of infection (Wong et al., 1983). All mountain lion sera tested were negative for the cuticular antigen but one serum was positive for the somatic antigen, a less specific test (Wong et al., 1983). This animal was negative for circulating microfilaria in whole blood. The low antibody prevalence may reflect lack of exposure, a less vigorous immune response than the domestic cat, or low sensitivity of the test when applied to the mountain lion.

Based on the detection of specific serum antibodies, we suggest prior exposure to that infectious agent or a related antigen. A single seropositive sample does not indicate ongoing disease nor previous morbidity. Only one animal had signs of clinical disease with a corresponding elevated FPLV titer. This combination of clinical disease and elevated titer support an etiological diagnosis but are not themselves diagnostic. Many serological tests may cross-react with several similar antigens,

and are not specific for a single infectious agent. Seroprevalence may reflect exposure to a feline pathogen or exposure to related antigens through prey items. The domestic cat and bobcat have been shown to be prey items of the mountain lion by stomach content analysis (D. A. Jessup, pers. comm.).

Other serological tests, such as FIV and FeLV viral core antigen assays, are used to detect the presence of the virus. Based on the 100% negative results from the sample population, we believe the virus is absent from healthy mountain lions. In combination with the lack of seroconversion to FeLV, the free-ranging mountain lion either may be highly vulnerable because of no previous exposure or resistant and non-responsive to these feline pathogens. Since FIV has been confirmed in the Florida free-ranging mountain lion population, it is more likely a matter of exposure, although the sensitivity of the tests used in this survey may be debated.

Serological surveys assess exposure to infectious agents in animal populations whose disease status is unknown. Seroprevalence to various feline pathogens reflects exposure to those agents but may not reflect the effect of these agents on the health of the free-ranging population. Antibody prevalence to zoonotic diseases such as *Yersinia pestis* and *Toxoplasma gondii* identify the mountain lion as a possible sentinel within the ecosystem.

Knowledge of potential disease exposure within the mountain lion population of California is important when evaluating ecological pressures affecting this species. As mountain lion numbers increase due to a ban on sport hunting and as population densities increase due to decreased habitat, the potential exists for these and other infectious agents to affect morbidity and mortality.

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