

## Prevalence of infectious diseases in feral cats in Northern Florida

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**Summary** Objectives of this study were to determine prevalence of infection in feral cats in Northern Florida with a select group of infectious organisms and to determine risk factors for infection. Blood samples or sera from 553 cats were tested with a panel of antibody, antigen or PCR assays. Male cats were at higher risk for FIV, *Mycoplasma haemofelis*, and *M. haemominutum*. Infection with either FeLV or FIV was associated with increased risk for coinfection with the other retrovirus, *M. haemofelis*, or *M. haemominutum*. *Bartonella henselae* had the highest prevalence and was the only organism that did not have any associated risk for coinfection with other organisms. Feral cats in this study had similar or lower prevalence rates of infections than those published for pet cats in the United States. Thus, feral cats assessed in this study appear to be of no greater risk to human beings or other cats than pet cats.

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## Introduction

The number of feral cats in the United States is unknown, but is suspected to rival the number of pet cats (73 million in 2002) (Levy et al., 2003). Issues of concern include the welfare of the cats themselves, public nuisances they may cause, their impact on the environment, and their impact on public health of both cats and humans. The National Association of State Public Health Veterinarians (NASPHV) states that "... the impact of these animals on human public health is defined by zoonotic diseases including rabies ... bartonellosis ... and toxoplasmosis (NASPHV Year 2000 Action Plan)." In addition, the NASPHV states, "There is no evidence that colony management programs reduce diseases." In contrast, The American Veterinary Medical Association accepts the maintenance of controlled, managed colonies of feral cats, as long as they are sterilized, identified, tested for infectious diseases and adopted or euthanized if positive (AVMA, 1996). These statements typify an ongoing debate regarding this population of cats. Despite this, there are few objective data regarding the actual prevalence of infectious diseases of feral cats in the United States.

Recent publications have shed light on various issues surrounding feral cats, including the demographics of the cats and their caretakers and the effects of Trap-Neuter-Return (TNR) programs on the overall population (Levy et al., 2003; Scott et al., 2002). The purpose of this study was to extend this information database and determine the prevalence and risk factors of infectious diseases in a population of feral cats. The term "feral" used in this study includes free-roaming stray and feral cats and implies a lack of confinement and ownership. The infectious organisms studied here were chosen because of their importance to feline or human health. Feline leukemia virus (FeLV), feline immunodeficiency virus (FIV) and feline coronavirus (FCoV) are organisms that spread directly from cat to cat; feral cats could serve as a source of infection to pet cats allowed outdoors or other free-roaming cats. *Dirofilaria immitis* (mosquitoes), *Mycoplasma haemofelis* (*Ctenocephalides felis*), *M. haemominutum* (*C. felis* suspected), *Bartonella henselae* (*C. felis*), *Ehrlichia* spp. (tick suspected), and *Anaplasma phagocytophilum* (*Ixodes* spp.) are infectious agents that cause clinical disease in some cats and are proven or suspected to be vector-borne. In addition, some *Ehrlichia* spp. and *A. phagocytophilum* can also infect human beings. Feral cats may play a role in spread of *Toxoplasma gondii* oocysts in the environment, indirectly

resulting in the infection of a variety of species including domestic cats and human beings. *Bartonella henselae* is a flea-borne zoonotic agent that occasionally causes clinical illness in cats; feral cats may play a role in magnifying the organism in *C. felis* that then infest pet cats and human beings.

## Materials and methods

### Animals

Cats were selected from those admitted to a TNR program (Operation Catnip<sup>®</sup>, Inc.) in Gainesville, Florida, from June 1999 to February 2000. Cats were anesthetized, surgically sterilized, vaccinated, and released back to the location where they were trapped. The tips of the left ears were trimmed to identify sterilized cats. Blood samples were collected specifically for this study from as many cats as possible without disrupting the operation of the clinic. No attempt was made to select cats based on gender or condition, and only cats judged to be adults based on the presence of permanent canine teeth and opinion of the surgeons were included in the study. Cats were presumed healthy based on observations of the caretakers, handlers and veterinarians at the TNR program. Thorough physical examinations and hematology were not routinely performed. A total of 553 cats (287 males and 266 females) were sampled.

### Sample collection

Blood samples obtained by jugular venipuncture were placed in serum separator and EDTA treated glass tubes. Serum and whole blood were stored at  $-80^{\circ}\text{C}$  until diagnostic testing was performed. Serum was obtained from 553 cats, while 484 (252 from males and 232 from females) whole blood samples were acquired.

### Testing methodology

#### FeLV antigen and FIV antibody

Serum was either shipped for batch testing by a commercial lab for the presence of FeLV p27 antigen (PetChek<sup>®</sup> FeLV Antigen Test; IDEXX) and FIV antibody (PetChek<sup>®</sup> FIV Antibody Test; IDEXX) by microtiter plate ELISA test kits ( $n=550$ ) or by using a commercially available in-hospital test kit (SNAP<sup>®</sup> FIV Antibody/FeLV Antigen Combo; IDEXX) ( $n=3$ ).

### Coronavirus antibody

FCoV antibody titer was determined by indirect immunofluorescence antibody assay (IFA) as described (Harpold et al., 1999). Titers > 1:40 were considered seroreactive.

### *Dirofilaria immitis* antibody and antigen

Samples were shipped to a commercial laboratory for *D. immitis* antigen and antibody testing using microtiter plate ELISA test kits (NOW™; Animal Diagnostics) as described (Watkins et al., 1998).

### *Toxoplasma gondii* antibody

IgM and IgG antibodies against *T. gondii* were determined by microtiter plate ELISA as described (Lappin and Powell, 1991). For both IgM and IgG antibody, titers = 1:64 were considered seroreactive.

### *Bartonella henselae* antibody

*Bartonella henselae* antibody titers were determined by IFA assay as described (Breitschwerdt et al., 1995). Titers > 1:64 were considered seroreactive.

### PCR assays

DNA was extracted from whole blood as previously described. PCR assays that amplify *M. haemofelis* DNA, *M. haemominutum* DNA, *Ehrlichia* spp. DNA, and *A. phagocytophilum* DNA were performed on each DNA sample (Brewer et al., 2003; Jensen et al., 2001).

### Statistical analysis

Presence of FeLV antigen, FIV antibody, *D. immitis* antigen, *M. haemofelis* DNA, *M. haemominutum* DNA, *Ehrlichia* spp. DNA, or *A. phagocytophilum* DNA in the blood or serum documents current infection. Presence of antibodies against FCoV,

*D. immitis*, *T. gondii*, and *B. henselae* could denote either current or previous infection. For the purpose of this study, we considered any positive test result to be evidence of infection.

Prevalence estimates were calculated for each organism and reported as the percent of cats with a positive test result (number of cats classified as positive divided by the total number of cats tested). In addition, odds ratios (OR) and 95% confidence intervals (95% CI) were calculated. In this study, the OR was used as an epidemiologic measure of association between exposure (e.g., gender) and risk of disease (e.g., FIV). Thus, if a particular exposure or risk factor was not associated with risk of disease, the OR was 1. The greater the departure of the OR from 1 (either larger or smaller), the stronger the association was between the factor and risk of disease. The upper and lower limits of a 95% CI indicate that one can be 95% confident in the assertion that the true OR falls within this interval. If the interval is broad, the precision is low. Proportions were compared using the chi-squared test. *P* values < 0.05 were considered statistically significant.

## Results

The most common infections in feral cats were *B. henselae* and FCoV, followed by *Mycoplasma* spp., *D. immitis*, and *T. gondii* (Table 1). Of the 101 (18.3%) cats that had antibodies to FCoV, most had low antibody titers with only 7 cats having titers of 1:320 or greater. Three of 553 cats (0.5%) were positive for both FeLV and FIV. All *D. immitis* Ag positive cats were *D. immitis* Ab positive, but only 7 of 64 (10.9%) *D. immitis* Ab positive cats were *D. immitis* Ag positive. Concurrent *M. haemofelis* and *M. haemominutum* infections were detected in 19 of 484 cats (3.9%). Of the 67

**Table 1** Prevalence of infections in feral cats in Northern Florida

Infectious organism	Percent positive <sup>a</sup>	Testing modality
FeLV	3.3 (18/553)	ELISA for antigen
FIV	5.2 (29/553)	ELISA for antibody
Feline coronavirus	18.3 (101/553)	IFA for antibody
<i>D. immitis</i>	11.6 (64/553)	ELISA for antibody
<i>D. immitis</i>	1.3 (7/553)	ELISA for antigen
<i>M. haemofelis</i>	8.3 (40/484)	PCR for DNA
<i>M. haemominutum</i>	12.2 (59/484)	PCR for DNA
<i>Ehrlichia</i> spp./ <i>A. phagocytophilum</i>	0 (0/484)	PCR for DNA
<i>T. gondii</i> IgM	2.0 (11/553)	ELISA for antibody
<i>T. gondii</i> IgG	8.9 (49/553)	ELISA for antibody
<i>B. henselae</i>	33.6 (186/553)	IFA for antibody

<sup>a</sup> Numbers in parentheses indicate number of cats with positive test results/Number of cats tested.

**Table 2** Risk of gender (male) associated with infectious organisms

Infectious organism	Odds ratio	95% confidence interval	P-value
FIV	4.76	1.80–12.57	0.001
<i>M. haemofelis</i>	6.13	2.94–12.79	<0.001
<i>M. haemominutum</i>	4.84	2.10–11.17	<0.001
<i>B. henselae</i>	0.64	0.45–0.92	0.01
<i>T. gondii</i> IgM	0.25	0.08–0.78	0.01

(12.1%) *T. gondii* seroreactive cats, 11 (2.0%) were positive for IgM alone, 49 (8.9%) were positive for IgG alone, and 7 (1.3%) were positive for both IgM and IgG.

Male cats were at significantly higher risk for FIV, *M. haemofelis* and *M. haemominutum*. Female cats were at slightly higher risk for *B. henselae* and *T. gondii* IgM (Table 2). Associations between the retroviruses and *Mycoplasma* spp. were the most noteworthy (Table 3). Infection with FeLV was associated with increased risk for coinfection with and detection of both FIV Ab and *M. haemominutum* DNA. Infection with FIV was associated with increased risk for coinfection with FeLV, *M. haemofelis*, and *M. haemominutum*. Cats infected with one species of *Mycoplasma* were at risk to be coinfecting with the other. Coronavirus was associated with FIV, *T. gondii* IgM, and *M. haemominutum*. Cats with an IgG titer for *T. gondii* were at increased risk for *D. immitis* Ab or Ag. *B. henselae* was the most common infection (33.6% prevalence) and was the only organism that did not have any associated risk for coinfection with other organisms.

## Discussion

Considerable debate is ongoing about feral cat populations in the United States. These cats have long been an area of concern in other countries, particularly European countries, as evidenced by the frequency of disease prevalence data available compared to the United States. Increased interest and awareness of TNR programs have brought feral cats to the forefront of animal control discussions in the United States. Aside from recently reported prevalences of FeLV and FIV by Lee et al. (2002), epidemiological information about infectious diseases, both feline and zoonotic, has yet to be reported in large feral cat populations in the United States.

FeLV and FIV are 2 of the most common infectious diseases of cats. Both viruses cause immunosuppressive, wasting syndromes in infected cats. The major routes of transmission for FeLV are via saliva through close casual contact (grooming), shared dishes or litter pans, and bite wounds and also by milk. The primary route of transmission of FIV is via bite wounds. Diagnosis is routinely made using ELISA and immunochromatographic tests for detection of soluble p27 FeLV antigen or FIV antibody in whole blood, serum or plasma.

Both FeLV and FIV have worldwide distribution. Prevalence of FeLV in pet cats ranges from 2% to 18% (Bandeccchi et al., 1992; Malik et al., 1997; Yamamoto et al., 1989); prevalence of FIV ranges from 1.2% to 43.9% (Bandeccchi et al., 1992; Fisch and Altman, 1989; Ishida et al., 1989; Malik et al., 1997; O'Connor et al., 1991; Yamamoto et al.,

**Table 3** Risk of disease (outcome) associated with infection with other organisms (exposure)

Outcome	Exposure	Odds ratio	95% confidence interval	P-value
FeLV	FIV	3.91	1.07–14.36	0.03
	<i>M. haemominutum</i>	3.19	1.08–9.38	0.03
FIV	Coronavirus	2.50	1.13–5.56	0.02
	<i>M. haemofelis</i>	10.71	4.67–24.56	<0.001
	<i>M. haemominutum</i>	4.74	1.86–12.10	0.001
Coronavirus	<i>T. gondii</i> IgM	2.99	1.13–7.89	0.02
	<i>M. haemominutum</i>	2.14	1.14–4.03	0.01
<i>D. immitis</i> Ab	<i>T. gondii</i> IgG	3.34	1.73–6.48	<0.001
	<i>D. immitis</i> Ag	59.8	7.5–476.3	<0.01
<i>D. immitis</i> Ag	<i>T. gondii</i> IgG	6.98	1.52–32.01	0.01
<i>M. haemominutum</i>	<i>M. haemofelis</i>	9.14	4.54–18.37	<0.001
<i>Ehrlichia</i> spp.	None	–	–	–
<i>T. gondii</i> IgM	<i>T. gondii</i> IgG	6.31	2.34–17.02	<0.001
<i>T. gondii</i> IgG	<i>T. gondii</i> IgM	6.31	2.34–16.99	<0.001
	<i>M. haemominutum</i>	2.48	1.18–5.18	0.01
<i>B. henselae</i> Ab	None	–	–	–

1989). Variations in reported prevalence rates are most likely due to regional variations in the populations tested, the health of the cats tested and whether cats were selected from high vs. low risk populations. Infection rates were highest (43.9%) in clinically ill cats ( $n=3323$ ) in a population sampled in Japan (Ishida et al., 1989). In the United States, most serosurveys are reported in sick and high-risk pet cats. In a national survey of sick and high-risk pet cats, prevalence of FeLV and FIV in Florida ( $n=2643$ ) was 12.8% and 8.4%, respectively (O'Connor et al., 1991). A smaller study of pet cats in southeastern Florida ( $n=95$ ) also reported an FIV prevalence of 8.4% (Fisch and Altman, 1989). In the current study, FeLV and FIV prevalences were relatively low (FeLV 3.3%, FIV 5.2%) when compared to infection rates reported for owned cats in the United States. Thus, feral cats do not appear to be a greater reservoir for these retroviruses than pet cats.

Historically, free-roaming, sexually intact male cats have been found to have the highest infection rate for FIV since they are more likely to be territorial and to be engaged in catfights (Ishida et al., 1989). The increased risk of *Mycoplasma* spp. in males and strong association between FIV and *Mycoplasma* spp. identified in the current study suggest that bite wounds may play a role in transmission of *Mycoplasma* spp. as well. The association of retrovirus infections with *Mycoplasma* infection also suggests that either the immunosuppressive effects of the retroviruses enhance *Mycoplasma* spp. as a secondary infection or that the risk factors are similar for both retroviruses and mycoplasmas. A recent study of pet cats ( $n=46$ ) in Israel also identified a correlation between retroviruses and detection of *Mycoplasma* spp. via blood smear and reported an increased morbidity in cats with coinfection (Harrus et al., 2002). Similar findings were documented in experimentally infected cats (George et al., 2002).

The clinical condition of feline infectious peritonitis (FIP) is caused by a pathogenic coronavirus. It manifests in a variety of clinical signs and can affect nearly any body system. It is thought that the FIP virus (FIPV) is the result of de novo mutations of the minimally pathogenic feline enteric coronavirus (FECV) (Vennema et al., 1998). The term feline coronavirus (FCoV) includes both viruses. The major route of transmission of FECV is via fecal shedding of the virus and subsequent oronasal contact. Establishing a diagnosis of FIP can be difficult, but commonly includes testing for antibodies to coronavirus. Positive tests are not diagnostic of disease, but rather only indicate exposure to FCoV. In fact, most cats with FCoV

antibody never develop FIP. A serologic survey in Davis, CA reported antibodies to FCoV in 20% of pet cats ( $n=33$ ) and 87% of purebred cats ( $n=108$ ) in catteries (Pedersen, 1976). A recent study of stray cats in Britain ( $n=517$ ) identified a prevalence of FCoV antibodies in 22.4% of cats (Muirden, 2002). In the current study, the prevalence of FCoV antibodies (18.3%) was lower than reported in pet cats and the antibody titers were low in most seroreactive cats. This suggests minimal exposure to FCoV in feral cats. Thus, feral cats in this population do not appear to be a greater source than pet cats for shedding of coronavirus in the environment. This might be due to the lower population density of feral cats compared to pet cats and their practice of burying feces outside in contrast to sharing a litter box. The presence of coronavirus antibodies was statistically associated with FIV, *T. gondii* IgM, and *M. haemominutum*, however, none of the relationships were very strong (Table 3). Oronasal contact with feces is a route of transmission for both *T. gondii* and FCoV, thus this could be an explanation for the association between these two organisms. However, one might have expected an association between FCoV and *T. gondii* IgG antibody as well. In a study of cattery and stray cats ( $n=275$ ) in Davis, CA, concurrent infections with multiple other infectious diseases were not found to be associated with increased risk for developing FIP (Foley et al., 1997).

Haemoplasmosis (formerly haemobartonellosis) is caused by *M. haemofelis* and *M. haemominutum*. *Haemobartonella felis* has undergone recent reclassification into the genus *Mycoplasma* based on analysis of 16S ribosomal ribonucleic acid gene sequences. *H. felis* large form (Ohio variant) and *H. felis* small form (California variant) have been renamed *M. haemofelis* and *M. haemominutum*, respectively (Foley and Pedersen, 2001; Neimarket et al., 2001, 2002; Rikihisa et al., 1997). In experimentally inoculated cats, increased morbidity is associated with *M. haemofelis* compared to *M. haemominutum* (Foley et al., 1998; Jensen et al., 2001; Westfall et al., 2001). *Mycoplasma haemominutum* has been detected in both naturally exposed cats and their fleas. It is taken into fleas during a blood meal, but transmission to naïve cats was not documented in a preliminary study of three cats (Woods et al., 2003). However, *M. haemofelis* was transmitted from an infected cat into a naïve cat by *C. felis* (Woods and Lappin, Unpublished data, 2003). These results suggest that *C. felis* may be a mode of transmission of these organisms between cats. Our findings that *Mycoplasma* spp. infection is more common in male cats and FIV-infected cats suggest that bite wounds may

play a role in transmission. Based on cytology, which cannot accurately differentiate *M. haemofelis* and *M. haemominutum*, worldwide prevalence of haemoplasmosis ranges from 3.6% to 42% of cats (Grindem et al., 1990; Harbutt, 1969; Nash and Bobade, 1986; Yamaguchi et al., 1996). The highest prevalence was in an isolated group of farm cats ( $n=48$ ) studied in Oxford, UK (Yamaguchi et al., 1996). In another study of pet cats ( $n=123$ ) in Wake County, NC, a prevalence rate of 4.6% was identified (Grindem et al., 1990). A third prevalence study was in pet cats ( $n=155$ ) at the University of Glasgow, Scotland, where the prevalence was 23.2% (Nash and Bobade, 1986). Genetically, *M. haemominutum* from the United States and United Kingdom are virtually identical (Tasker et al., 2001). A recent report surveyed 426 pet cats using a PCR assay at the University of Bristol, UK and identified a prevalence of 1.4% and 16.9% for *M. haemofelis* and *M. haemominutum*, respectively (Tasker et al., 2003a,b). In 220 pet cats in the United States surveyed with a PCR assay, 4.5% were infected with *M. haemofelis*, 12.7% with *M. haemominutum*, and 2.3% were infected with both *Mycoplasma* spp. giving an overall prevalence of 19.5% (Jensen et al., 2001). These studies demonstrate considerable variation in percent prevalence depending on the population studied and the technique used to establish infection. The prevalence in the current study falls within the lower end of the reported ranges for pet cats, thus, feral cats do not serve as a greater reservoir of mycoplasmas than pet cats. In the current study, cats infected with one *Mycoplasma* species were at increased risk to be infected with the other, which supports the hypothesis that the route of transmission for both may be similar. Associations were found between FeLV, FIV and *Mycoplasma* spp. as well. There is no clear explanation for the associations between *Mycoplasma* spp. and FCoV or *T. gondii* based on the known biologic behavior of these agents.

*Dirofilaria immitis* is a parasite that primarily affects dogs, however, diagnosis in cats is on the rise (Atkins et al., 1998, 2000; Kalkstein et al., 2000; Snyder et al., 2000). This challenging diagnosis is made based on clinical signs, radiographic and echocardiographic findings, and serologic testing. Tests are available for both Ag and Ab (Goodwin, 1998; Prieto et al., 1997), however, neither has proven to be solely efficacious for definitive diagnosis. The presence of antibody does not necessarily indicate a current infection, only exposure at some previous time. Antigen tests detect proteins from mature female worms. Low worm burden and male-only heartworm infections in cats reduce the quantity and type of Ag available for

detection, resulting in false-negative Ag test results. In a recent study evaluating the efficacy of serologic tests for the detection of heartworm, Ag tests detected 79–86% of heartworm infections and were highly specific, whereas Ab tests were less sensitive, detecting 62–72% of heartworm infections, and had a wider range of false-positive results than Ag tests (Berdoulay et al., 2002). Infections in cats diagnosed mostly by necropsy in the United States have been reported in 38 of the 50 states with a prevalence of 0 to 14% (Atkins et al., 2000; Hermesmeier et al., 2000; Kalkstein et al., 2000; Kendall et al., 1991; Labarthe et al., 1997; McCall et al., 1994; Willard et al., 1988). A prevalence study of pet cats ( $n=7969$ ) in the United States identified Ab and Ag prevalence in Florida to be 22.5% and 4.8%, respectively (Watkins et al., 1998). The prevalence of infection in the current study is lower than the previous report from this region. Consequently, this population of cats does not appear to be at greater risk of *D. immitis* infection when compared to pet cats. All of the Ag positive cats in this study were also Ab positive, but only 10.9% of Ab positive cats were Ag positive.

Experimentally, cats have been infected with *Neorickettsia risticii* (previously *E. risticii*) and *A. phagocytophilum* (previously *E. equi*) (Dawson et al., 1988; Dumler et al., 2001; Lewis et al., 1975; Neer et al., 2002). Naturally occurring ehrlichial infections in cats have been documented via antibody assays with seroprevalences ranging from 12% to 82.4% depending on location and infecting species (Bouloy et al., 1994; Dawson et al., 1988; Peavy et al., 1997; Perry et al., 1989). A national seroprevalence study of cats identified antibodies against *E. canis* and *N. risticii* in 29.2% of 583 cats tested with the predominant species being *N. risticii* (28.5%) (Stubbs et al., 2000). Although these studies report seroprevalences based on antibody testing, the relationship between antibodies and infection with ehrlichiosis or anaplasmosis in cats remains to be determined (Breitschwerdt et al., 2002; Neer et al., 2002). Some pet cats in the United States have been shown by PCR testing and genetic sequencing to be infected by an *E. canis*-like organism and *A. phagocytophilum* (Breitschwerdt et al., 2002; Lappin et al., 2003). Cats in this study were tested using PCR primers that can amplify *Ehrlichia*, *Anaplasma* and *Neorickettsia* spp (Brewer et al., 2003). As no amplification products were obtained from any of the cats surveyed in the current study, species-specific PCR tests which are also available were not utilized in this study (Breitschwerdt et al., 1998). It is likely that *A. phagocytophilum* is transmitted by

*Ixodes* spp. ticks which are uncommon in Florida (Akucewich et al., 2002). The failure to detect infections in this population of feral cats suggests that they are not associated with infection of pet cats.

*Toxoplasma gondii* is an obligate intracellular protozoan parasite. Felids serve as definitive hosts, transiently passing oocysts into the environment. Virtually all warm blooded animals serve as intermediate hosts. Infection is usually acquired transplacentally, by ingestion of tissue cysts or by ingestion of sporulated oocysts. In humans, *T. gondii* is of particular risk to pregnant women, with up to 50% of infections during pregnancy transmitted to the fetus (Jones et al., 2001). Since most cats and people do not clear the organism from tissues once infected, presence of IgM or IgG antibodies generally denotes infection (Lappin, 1996). However, since the oocyst shedding period is short, most seroreactive cats have completed the oocyst shedding period. Previously infected cats can occasionally repeat oocyst shedding, but the numbers are generally low and are not likely a significant public health risk. Serologic prevalence data indicate that toxoplasmosis is one of the most common infections of cats and humans throughout the world (Dubey, 1973; Dubey et al., 2002; Hill et al., 2000; Jones et al., 2001). Approximately 30% of pet cats and 22% of humans in the United States have *T. gondii* antibodies (Dubey, 1973; Hill et al., 2000; Jones et al., 2001). A recent study of pet cats ( $n=275$ ) in rural Ohio identified a prevalence of 48% (Dubey et al., 2002). Another study of cats ( $n=205$ ) from Colorado found a prevalence of 19.7% in pet cats and 29.8% in cats from a humane shelter (Hill et al., 2000). In the only national seroprevalence study of clinically ill pet cats in the United States ( $n=12,628$ ) an overall prevalence of 31.6% was documented, with 37.4% in the southeastern region of the United States (Voltaire et al., 2003). The prevalence observed in the current study (2.0% IgM, 8.9% IgG, 1.3% IgM + IgG) was lower than that reported for pet cats. Thus, feral cats do not appear to be more likely to shed the organism into the human environment than pet cats. It has been hypothesized that feral cats may be more likely than pet cats to ingest intermediate hosts harboring tissue cysts, however, there is no association between *T. gondii* prevalence in feral cats and wild rodents (DeFeo et al., 2002). Although previous studies have recognized an association between infections with FIV and *T. gondii* (Witt et al., 1989), no association was observed in this study. Based on the known biologic behavior of these agents, there is no clear explanation for the associations between *T. gondii* IgG antibodies and *D. immitis* or *M. haemominutum*.

*Bartonella henselae*, the organism that causes cat scratch disease (CSD) in humans, is a curved gram-negative bacillus that infects multiple species, including cats. The domestic cat is considered the reservoir for this organism. Infected cats can remain bacteremic for months to years. The pathogenicity of *B. henselae* in cats remains undetermined, however, most infected cats appear healthy (Breitschwerdt and Kordick, 2000). The organism is associated with multiple human diseases, including CSD, bacillary angiomatosis, visceral peliosis, bacteremia and endocarditis (Adal et al., 1994; Tompkins, 1997). These conditions can be life threatening in immunosuppressed humans. The cat flea, *C. felis*, is the primary vector of *Bartonella* spp. between cats (Chomel et al., 1996). Diagnosis is commonly made via IFA for antibodies to the organism. Alternative diagnostic tests include blood culture (Brenner et al., 1997) and PCR (Jensen et al., 2000). A serologic study of pet cats ( $n=628$ ) throughout the United States and in some areas of Canada identified an overall seroprevalence of 27.9% (range 3.7% to 54.6%) (Jameson et al., 1995). Seroprevalence was highest in warm, humid areas where flea infestation is more likely. A study of both pet and feral cats in Baltimore, MD found an association between seroreactivity to *B. henselae* and *T. gondii* (Childs et al., 1994). Although *B. henselae* was the most common infection identified in this study (33.6%), it was the only organism that did not have any associated risks for coinfection with other organisms. Female cats were at slightly greater risk for being infected than males. Flea infestations have been identified in 92% of feral cats in Florida (Akucewich et al., 2002), thus grooming and ingestion of fleas could explain an increased rate of infection, particularly females grooming flea infested kittens. Exposure to kittens with fleas is the highest risk factor for CSD in humans as well (Zangwill et al., 1993).

The results of this study should be interpreted with some caution. The samples were collected from feral cats that were trapped by caretakers for the purposes of neutering. Thus, the tested population is skewed toward feral cats that have some association with humans and is not a random sample of feral cats. However, since feral cat interaction with humans is a primary issue that concerns public health officials, this is an appropriate population of cats to test. Some testing techniques, such as heartworm antibody, indicate only exposure to infection, but not necessarily current infection. This is an unavoidable limitation of the current assays available, and is a problem also encountered in the diagnosis of infections in pet cats. We did not test cats for all of the important

infectious diseases of public health concern. The most noticeable omission is rabies, for which there are no accurate ante-mortem tests. In addition, we did not test for internal or external parasites or enteropathogens.

Feral cats in this study had similar prevalence of infections compared to those published for pet cats in the United States. This suggests that feral cats assessed in this study appear to be of no greater risk to human beings or other cats than pet cats.

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