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Seroprevalence of *Bartonella henselae* Infection and Correlation with Disease Status in Cats in Switzerland

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The prevalence of infection with *Bartonella henselae* was investigated in cats from different areas of Switzerland. Serum samples of 728 cats were examined for antibodies to *B. henselae* by immunofluorescent antibody testing, and the results were analyzed with a view to a possible correlation between a positive titer and signalment, clinical signs, infection with feline leukemia virus (FeLV), feline immunodeficiency virus (FIV), feline coronavirus (FCoV), or feline spumavirus (FeSFV), and the living environments of the cats. The seroprevalence in all cats was 8.3%. No significantly different prevalence was found in sick versus healthy cats (9.2 versus 7.2%); however, in sick cats seropositive for *B. henselae*, there was an increased frequency of stomatitis and a variety of diseases of the kidneys and the urinary tract. There was an increased prevalence of *B. henselae* in cats positive for FCoV ($P = 0.0185$) or FeSFV ($P = 0.0235$) and no statistically significant increased prevalence in cats infected with FeLV or FIV. There was no correlation between a positive titer and sex or breed. The same prevalence of *B. henselae* antibodies was found in cats with and without access to the outdoors and in cats from single- and multicat households. The seroprevalence was increased in cats living south of the Alps (12.1%); however, this difference was not significant ($P = 0.0616$).

Bartonella henselae has been demonstrated to be a causative agent of cat scratch disease (6, 21, 27), bacillary angiomatosis (13), endocarditis (8), bacteremia, and hepatic peliosis (26) in humans. Cat bite or scratch is strongly associated with cat scratch disease (27) and bacillary angiomatosis (24). Bartonellosis has been diagnosed worldwide; however, its prevalence, and therefore its impact, differs among geographic regions. In warm, humid areas, the seroprevalence in cats has been shown to be much higher than in cold areas, and an association with the presence of fleas has been made (11).

However, few data are available concerning the clinical importance of *B. henselae* for cats, and no data are available concerning the prevalence of *B. henselae* in cats in Switzerland. The aim of this study was to investigate whether seropositivity was associated with the occurrence of viral infections, with clinical disease, or with the signalment and living conditions of the cats, and to document the prevalence of *B. henselae* in cats in different parts of Switzerland and southern Germany on the basis of serological examination.

MATERIALS AND METHODS

Between 1987 and 1990, serum samples from 728 cats (304 healthy, 424 sick) were collected from veterinary practices in different regions in Switzerland and southern Germany. Information with respect to signalment, disease history, abnormal clinical findings, and environment was gained from the participating veterinarians by use of a questionnaire. Serum samples were evaluated for the presence of antibodies to *B. henselae*, feline immunodeficiency virus (FIV), feline coronavirus (FCoV), feline spumavirus (FeSFV), and feline leukemia virus (FeLV) antigen. Antibodies to *B. henselae* were detected by an immunofluorescence antibody test (IFA) using a technique described previously (21). Briefly, Vero cells grown in RPMI 1640 medium supplemented with fetal bovine serum were infected with *B. henselae*. Infected cells were adjusted to 50,000/ml, and 50-μl aliquots were spotted onto glass slides, air dried, and fixed in acetone.

Positive- and negative-control sera were used in a dilution of 1:50 with every slide. Cat serum samples were incubated at different dilutions at 37°C for 1 h, followed by a washing step and incubation with a goat anti-cat immunoglobulin G conjugated to fluorescein isothiocyanate (Jackson ImmunoResearch Laboratories, Inc.; distributed by Milan AG, La Roche, Switzerland). The IFA reaction was considered positive if specific fluorescence was noted at a titer of ≥64. Serum samples showing fluorescence at a 1:64 dilution were titrated by twofold dilutions up to 1:8,192.

FeLV p27 antigen was demonstrated by a double-antibody sandwich enzyme-linked immunosorbent assay (ELISA) as described previously (18). Antibodies to FCoV and FeSFV were measured by IFA as described previously (14, 16). Antibodies to FIV were detected by ELISA. Positive results were confirmed by the Western blot technique (15). Serum samples were kept frozen at −70°C until assayed. Data from *B. henselae* serology were examined with respect to health status, prevalence of diseased organ systems, breed, age, sex, living environment (indoors versus outdoors; single- versus multicat household), and coinfection with FeLV, FCoV, FIV, or FeSFV. Frequencies were compared by Fisher's exact test. Differences were considered significant if P was <0.05.

RESULTS

Exact data concerning FIV, FCoV, and FeLV are compiled elsewhere (17). The overall prevalence of antibodies to *B. henselae* was found to be 8.3% (61 of 728 samples analyzed). No significant difference was found in seroprevalence between healthy and sick animals. Of 304 healthy cats tested, 22 were seropositive (7.2%). Their median titer was 128 (range, 64 to 8,192). Of 424 sick cats tested, 39 were seropositive (9.2%). Their median titer was 128 (range, 64 to 2,048). On the basis of the questionnaires, about 30 parameters were evaluated for each cat and compared with respect to *B. henselae* serostatus. Upon physical examination, two abnormalities were found at an increased frequency in sick cats seropositive for *B. henselae*: stomatitis ($P = 0.0117$) and various diseases of the kidneys and urinary tract that were not further differentiated ($P = 0.0337$).

Cats seropositive for *B. henselae* were more often seropositive for FCoV (26 of 61 *B. henselae*-seropositive cats versus 186 of 667 *B. henselae*-seronegative cats; $P = 0.0185$) and FeSFV ($P = 0.00235$) (data not shown). Sick cats seropositive for *B. henselae* were more often infected with FIV; however, this difference was not significant ($P = 0.0616$). The prevalence of seropositive cats was not influenced by infection with FeLV

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TABLE 1. Seroprevalence of *B. henselae* in healthy cats in relation to feline viral infections

Virus and titer	No. (%) of healthy cats (n = 304)		No. (%) of sick cats (n = 424)	
	B. henselae positive (n = 22)	*B. henselae* negative (n = 282)	*B. henselae* positive (n = 39)	*B. henselae* negative (n = 385)
FCoV				
<1:25	15 (68.2)	226 (80.1)	20 (51.3)	255 (66.2)
1:25	4 (18.2)	28 (9.9)	10 (25.6)	66 (17.1)
1:100	2 (9.1)	21 (7.4)	7 (17.9)	36 (9.3)
1:400	1 (4.5)	7 (2.5)	2 (5.1)	19 (4.9)
1:1,600	0	0	0	9 (2.3)
≥1:25	7 (31.8)	56 (19.9)	19 (48.7)	130 (33.8)
FIV positive[a]	0	4 (1.4)	3 (7.7)	9 (2.3)
FeLV p27 positive	1 (4.5)	2 (0.7)	2 (5.1)	46 (11.9)
FeSFV positive[b]	12/20 (60.0)	92/274 (33.6)	20/35 (57.1)	176/362 (48.6)

[a] By ELISA and Western blot.
[b] By IFA; cutoff, ≥1:40.

(Table 1). There was no correlation between privately owned cats versus cats brought to an animal shelter, indoor versus outdoor cats, single- versus multicat households, sex, or breed and a titer positive for *B. henselae*. In cats up to the age of 7 years, there was no correlation between age and a positive titer; however, above the age of 7 years, more cats were seropositive (17 of 110; $P = 0.0077$) (Fig. 1), and sick cats more often were seropositive (17 of 93; $P = 0.00235$). In fact, all the seropositive cats in this age group were sick.

There was a difference in prevalence with respect to the geographic region. Cats living south of the Alps (Tessin) had a higher prevalence (12.1%; 16 of 132) than animals inhabiting the northern parts of Switzerland and Germany; however, this difference was not significant ($P = 0.0616$).

DISCUSSION

The data presented here indicate that cats in Switzerland are fairly commonly infected with *B. henselae*. This prevalence of 8.3% is markedly lower than those found in other studies of cats in other countries (1, 2, 4). Climatic differences among these countries are one possible explanation for this difference. Jameson et al. (11) have shown that the prevalence of positive titers is affected by climatic warmth and annual precipitation. Switzerland is climatically comparable to the northern United States and Canada, where the prevalence is low (11). We also have recognized a higher prevalence south of the Alps, although this difference was not significant. A higher prevalence in the south can be explained by the fact that average temperatures south of the Alps are considerably higher. The average temperature north of the Alps is 3.5°C lower (23). Increased average temperatures lead to longer survival of fleas, which in turn can explain a higher infectious pressure on cats by *B. henselae.*

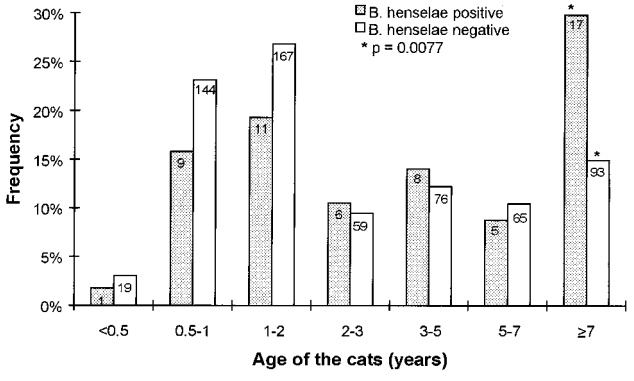

FIG. 1. Age distribution (frequency of a particular age in percentages) of *B. henselae*-positive and -negative cats. The ages of 48 cats were unknown. Numbers in the bars are the numbers of animals per group. Cats that were ≥7 years old were significantly more frequently *B. henselae* positive (*, $P = 0.0077$).

Another explanation for the low prevalence observed in this study may be an underestimation of the true prevalence of the infection. Underestimation may be expected if the bacteremic phase, which may last for months, is not paralleled by seroconversion. If this was the case, some of the acutely bacteremic cats would have been considered negative. However, in the study by Koehler et al. (12), there was a close relationship between bacteremic and seropositive cats, and it has been found experimentally that seroconversion follows shortly after, and lasts longer than, bacteremia (22). Therefore, it is likely that our study did not greatly underestimate the prevalence.

It is recognized that cats infected with *B. henselae* may be clinically normal. Although in our study seropositive cats were not sick more often than seronegative cats, an increased prevalence of stomatitis and various urinary tract abnormalities was found in seropositive cats. Seropositive cats also had a higher incidence of a positive titer for FeSFV and FCoV. An increased incidence of stomatitis in FeSFV-positive cats (14) and of renal abnormalities in FIV-positive cats (10) has been recognized. Hence, the increased prevalence of these clinical abnormalities in cats infected with *B. henselae* in our study may be associated with *B. henselae* infection, with one of the viral infections, or with a combination of these.

Several explanations for an increased prevalence of FCoV and FeSFV antibodies in *B. henselae*-positive cats are possible. First, there may be a problem in the methodology of the tests, e.g., cross-reactions in the IFA. This explanation is unlikely, because there were many cats seropositive for only one of the three diseases. Second, the correlation may be due to simultaneous transmission of these diseases. FCoV is most efficiently transmitted among cats living in close contact through the oropharyngeal route (20). FeSFV is transmitted through cat bites (19). Although *B. henselae* may be transmitted by bites or scratches, the main route of transmission appears to be through fleas (3, 5). Consequently, although transmission of these organisms does not occur by exactly the same means, transmission requires more or less close social contact. Alternatively, by immunosuppression one of these infections may

predispose to infection with, or delay clearing of, another organism. However, there was no significant correlation between viral diseases known to cause immunosuppression (FeLV and FIV) and *B. henselae* infection in the present study. Finally, a statistical correlation between diseases does not necessarily reflect a cause-and-effect relationship.

In contrast with the findings of Ueno et al. (25), clinical signs of cats seropositive for FIV and *B. henselae* did not differ from those of cats infected with FIV alone. This finding and the fact that the increased prevalence of FIV-positive cats was not significant may be associated with the low prevalence of FIV-positive cats in our population (2.2%).

Whereas, in contrast with the findings of other studies, no increased seroprevalence of *B. henselae* was seen in young cats, a statistical correlation with old age (≥7 years) was found. Assuming that, at least in some animals, seroreversion follows clearance of the infection (7), an explanation for this finding is that very old cats may be less immunocompetent and thus unable to clear the infection. Many of these cats were between 7 and 9 years old, making age-related immunosuppression an unlikely cause. On the other hand, all the seropositive cats in this age group were sick. Two explanations for this finding are possible: either *B. henselae* infection makes cats vulnerable to other diseases, or a diseased cat is less able to clear the infection with *B. henselae.*

It is expected that cats from multicat households will have a higher prevalence of *B. henselae* infection, as they are exposed to the same environment and the same vectors that may transmit the organism (3). However, crowding of cats did not play a role in the present study. It is also noteworthy that stray cats tested in animal shelters did not have an increased seroprevalence. Again, it is possible that more of these cats were infected than was recognized serologically. These cats may be bacteremic with *B. henselae* yet may not have seroconverted. Yet the main cause for this finding and the overall low prevalence in this study may be a very low prevalence of flea infestation in the cats in the tested geographic region.

In view of the prevalence of *B. henselae* infection among cats, the question arises whether immunosuppressed people should avoid all contact with cats. Considering the importance of the animal bond for human well-being, separation from an animal may not be wise. It may be advisable to screen pets owned by immunocompromised people for *B. henselae* infection and to treat positive animals. However, the optimal antibiotic regimen for clearing bacteremia in infected cats has yet to be found; so far, antibiotic treatment has not been shown to consistently clear the infection from cats (7). When the bacteremia has subsided, cats appear to be immune to reinfection with *B. henselae* (7, 22). It is at least as important to prevent and treat flea infestation in cats, as fleas have been shown to be a vector of the disease (5, 9).

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