

Seroprevalence of feline leukemia virus and feline immunodeficiency virus infection among cats in Canada

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Abstract – The purposes of this study were to determine the seroprevalence of feline leukemia virus (FeLV) and feline immunodeficiency virus (FIV) infection among cats in Canada and to identify risk factors for seropositivity. Signalment, lifestyle factors, and test results for FeLV antigen and FIV antibody were analyzed for 11 144 cats from the 10 Canadian provinces. Seroprevalence for FIV antibody was 4.3% and seroprevalence for FeLV antigen was 3.4%. Fifty-eight cats (0.5%) were seropositive for both viruses. Seroprevalence varied geographically. Factors such as age, gender, health status, and lifestyle were significantly associated with risk of FeLV and FIV seropositivity. The results suggest that cats in Canada are at risk of retrovirus infection and support current recommendations that the retrovirus status of all cats should be known.

Résumé – **Séroprévalence de l'infection par le virus de la leucémie féline et le virus de l'immunodéficience féline chez les chats au Canada.** Les buts de cette étude étaient de déterminer la séroprévalence du virus de la leucémie féline (FeLV) et du virus de l'immunodéficience féline (FIV) chez les chats au Canada et d'identifier les facteurs de séropositivité. Le signalement, les facteurs de style de vie et les résultats de tests pour l'antigène du FeLV et les anticorps du FIV ont été analysés pour 11 144 chats provenant de 10 provinces canadiennes. La séroprévalence pour l'anticorps du FIV était de 4,3 % et la séroprévalence pour l'antigène du FeLV était de 3,4 %. Cinquante-huit chats (0,5 %) étaient séropositifs pour les deux virus. La séroprévalence variait selon la géographie. Les facteurs comme l'âge, le sexe, l'état de santé et le style de vie étaient significativement associés au risque de séropositivité pour le FeLV et le FIV. Les résultats suggèrent que les chats au Canada sont à risque d'infection par les rétrovirus et appuient les recommandations actuelles que le statut rétroviral de tous les chats devrait être connu.

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Introduction

Feline leukemia virus (FeLV) and feline immunodeficiency virus (FIV) are retroviruses that represent 2 of the most common and important infectious diseases of cats worldwide. The American Association of Feline Practitioners (AAFP) recommends that the retrovirus status of all cats should be known and has published guidelines for retrovirus testing and management (1).

Several studies have evaluated the seroprevalence of FeLV and FIV infection in North American cats, but most of the

available data is for the United States. An earlier study evaluated FIV seroprevalence in 2254 high-risk cats and 511 cats of low or unknown risk from the United States and Canada (2). Of the high-risk cats, 42 were reported to be from Canada, but no specific location was given. Feline immunodeficiency virus seroprevalence was 19% (8/42) in this group of cats. Of the low or unknown risk cats, 352 were reported to be from Canada with no specific location indicated. The FIV seroprevalence was reported for the group as a whole (1.2%, 6/511) with no breakdown between cats from Canada versus those from the United States.

A recent study of 18 038 cats tested at North American veterinary clinics and animal shelters found 2.3% of cats seropositive for FeLV antigen and 2.5% seropositive for FIV antibody (3). Data on 325 cats from 7 unidentified Canadian provinces were included, with 2.5% of cats being seropositive for FeLV antigen and 3.1% being seropositive for FIV antibody. Three other studies have evaluated seroprevalence of FeLV and FIV in specific populations of Canadian cats. In 1 study, seroprevalence was evaluated in 246 cats from 3 demographic populations in Ottawa, Ontario (4). Seroprevalence for FIV antibody was highest in urban stray cats (23%; 17/74) and lower in client-owned cats (5.9%; 9/152) and in a feral cat colony (5%; 1/20).

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Table 1. Study population characteristics

Factor	Category	Samples		FIV+ test results		FeLV+ test results	
		Number	Proportion	Number	Prevalence	Number	Prevalence
Test site	Shelter/Rescue	1556	13.96	100	6.43	42	2.70
	Veterinary clinic	9588	86.04	380	3.96	341	3.56
Outdoor access	Yes	6532	58.61	339	5.19	258	3.95
	No	3607	32.37	69	1.91	84	2.33
	Unknown	1005	9.02	72	7.16	41	4.08
Age	Juvenile	4030	36.16	63	1.56	68	1.69
	Adult	7114	63.84	417	5.86	315	4.43
Gender	Spayed female	2423	21.74	58	2.39	100	4.13
	Intact female	2715	24.36	59	2.17	73	2.69
	Castrated male	3093	27.75	177	5.72	112	3.62
	Intact male	2873	25.79	183	6.37	95	3.31
	Unknown	40	0.36	3	7.50	3	7.50
FIV test result	Positive	480	4.31	NA	NA	58	12.08
	Negative	10 664	95.69	NA	NA	325	3.05
FeLV test result	Positive	383	3.44	58	15.14	NA	NA
	Negative	10 761	96.56	422	3.92	NA	NA
Health status	Healthy	7200	64.61	232	3.22	143	1.99
	Sick	3432	30.80	229	6.67	227	6.61
	Unknown	512	4.59	19	3.71	13	2.54
Province	Alberta	829	7.44	37	4.46	21	2.53
	British Columbia	1388	12.46	39	2.81	31	2.23
	Manitoba	656	5.89	19	2.90	39	5.95
	New Brunswick	120	1.08	1	0.83	0	0.00
	Newfoundland	300	2.69	15	5.00	13	4.33
	Nova Scotia	603	5.41	15	2.49	40	6.63
	Ontario	5613	50.37	216	3.85	144	2.57
	Prince Edward Island	187	1.68	7	3.74	8	4.28
	Quebec	1266	11.36	118	9.32	83	6.56
Saskatchewan	182	1.63	13	7.14	4	2.20	

NA — not applicable.

Seroprevalence for FeLV antigen was highest in urban stray cats (6.7%; 5/74) and lower in client-owned cats (2.6%; 4/152). No cats in the feral colony were seropositive for FeLV antigen. Two male cats (0.8%; 2/246) were seropositive for both FeLV and FIV infection.

In a study of a trap, neuter, and release program for feral cats on Prince Edward Island, 7.6% (14/185) of cats were seropositive for FIV antibody and 6.5% (12/185) were seropositive for FeLV antigen (5). Three male cats (1.6%) were seropositive for both FeLV and FIV. The seroprevalence of FIV was also evaluated in submissions of feline sera to a diagnostic laboratory in Atlantic Canada (6). Although 671 samples were tested, 90.5% were from Prince Edward Island. Seroprevalence of FIV antibody was 7.6%. The study identified an increasing risk of FIV seropositivity with increasing age, and found the prevalence of FIV antibodies was significantly higher in intact male cats than in other gender categories.

More information is needed on the seroprevalence of FeLV and FIV infection in cats in Canada to better define prophylactic, management, and therapeutic measures for owned and shelter cats. The purposes of this study were to determine seroprevalence of FeLV antigen and FIV antibody among cats from all 10 Canadian provinces and to identify risk factors for seropositivity.

Materials and methods

Study participants and population

Veterinary clinics, animal shelters, cat rescue programs, and feral cat programs in Canada were invited to participate in the study. Potential study participants were identified as all those who had purchased combination FeLV antigen and FIV antibody test kits or submitted samples for retrovirus testing to a diagnostic laboratory (Vita-Tech, Markham, Ontario; Montreal, Quebec; Central Laboratory for Veterinarians, Calgary, Alberta; Edmonton, Alberta; Langley, British Columbia) in the previous 12 mo. Potential participants were sent a letter explaining the study and inviting them to participate. An incentive was offered to participants who submitted a minimum of 15 complete test results before the end of the study period. Enrolled participants were asked to submit results of tests for FeLV and FIV performed between August 1, 2007 and November 15, 2007. Participants were requested to offer retrovirus testing to the owners of all cats and kittens in accordance with testing guidelines developed by the AAFP. The AAFP guidelines were provided to study participants.

Data collection

Information requested on each cat included test date, patient name, whether the test was performed in a clinic or at a referral

Table 2. Relationship of FIV and FeLV-seropositive test results with risk variables

Factor	Category	FIV			FeLV				
		Probability of positive test (%)	95% CI	<i>P</i> -value	Probability of positive test (%)	95% CI	<i>P</i> -value		
Test site	Shelter/Rescue	3.35	1.56–7.08	0.619	5.39	3.07–9.30	0.023		
	Veterinary clinic	2.83	1.70–4.68		9.17	6.27–13.22			
Outdoor access	Yes	5.00	2.88–8.54	< 0.001	8.19	5.37–12.31	0.027		
	No	1.46	0.75–2.81		5.85	3.63–9.30			
	Unknown	3.98	1.10–7.76		7.30	4.38–11.93			
Age	Juvenile	1.51	0.78–2.90	< 0.001	5.08	6.54–14.17	< 0.001		
	Adult	6.20	3.71–10.19		9.70	3.13–8.14			
Gender	Spayed female	1.40	0.78–2.49	0.541	5.52	3.61–8.35	0.485		
	Intact female	2.10	1.15–3.81		7.29	4.86–10.80			
	Castrated male	3.01	1.77–5.05		< 0.001	4.35		2.87–6.54	0.017
	Intact male	7.43	4.58–11.81			7.08		4.84–10.26	
	Unknown	4.13	0.95–16.26			13.62		4.03–37.22	
FIV test result	Positive	NA	NA	NA	10.97	6.85–17.14	< 0.001		
	Negative	NA	NA		4.46	2.97–6.64			
FeLV test result	Positive	4.88	2.67–8.77	< 0.001	NA	NA	NA		
	Negative	1.93	1.11–3.33		NA	NA			
Health	Healthy	2.21	1.24–3.92	< 0.001	4.26	2.75–6.54	< 0.001		
	Sick	4.72	2.63–8.36		12.72	8.35–18.90			
	Unknown	2.79	1.23–6.21		6.31	3.25–11.89			

NA — not applicable.

laboratory, age, gender, access to outdoors, and whether the cat was currently ill. Test results and information were submitted to the investigators by fax transmission using a standard reporting form.

Testing protocol

Cats were tested for FeLV antigen and FIV antibody using a commercially available point-of-care ELISA (SNAP Combo FeLV antigen/FIV antibody, IDEXX Laboratories, Westbrook, Maine, USA), or samples were submitted for enzyme-linked immunosorbent assay (ELISA) testing (PetChek FIV Antibody, PetChek FeLV Antigen; IDEXX Laboratories) to a diagnostic laboratory. The assay procedure was included in the package insert for each point-of-care test kit. Tests were performed on blood, plasma, or serum. Confirmatory testing was not performed as part of the study.

Risk factors

To evaluate regional variations in seroprevalence, data were grouped by the province of origin. Other risk factors that were evaluated included age [juvenile (< 6 months old) versus adult], gender (intact female, spayed female, intact male, castrated male), and whether cats had access to the outdoors. General health status of the cat at the time of testing was recorded as presence or absence of current illness.

Statistical analysis

Seroprevalence was determined by the percentage of cats with positive test results. Unadjusted seroprevalence estimates of FeLV infection, FIV infection, and FeLV/FIV co-infection were calculated for the study population as a whole and for each of the 10 provinces. A generalized linear mixed model was fitted

(SAS 9.1.3, Proc GLIMMIX; SAS, Cary, North Carolina, USA) to the binary outcome variables FIV or FeLV seropositive result. Effectively, a modified “logistic regression” model was established, with nominal explanatory variables and random effects. Practices, nested within shelter type and province, were treated as random effects (variance component). Fixed effects included the kind of test site (shelter or veterinary clinic), outdoor access, age category (juvenile or adult), health status, gender and province. Only practices with at least 1 case were included because practices with no cases are not informative with respect to the relation between any of the fixed effects and the outcome. A model was fitted to examine the main features of relationships. Cats used in the analyses only appeared once. Mean probabilities and odds ratios with approximate 95% confidence intervals (CI) were calculated for each effect. Significance was set at $P \leq 0.05$.

Results

A total of 343 veterinary clinics and 13 animal shelters or rescue organizations representing the 10 Canadian provinces participated in the study. Complete FeLV and FIV test results were received for 11 144 cats. More cats were tested by veterinary clinics ($n = 9588$) than by animal shelters/rescue organizations ($n = 1556$). Results are summarized in Tables 1 and 2.

Of the 11 144 cats with complete test data, 480 (4.31%) were seropositive for FIV antibody and 383 (3.44%) were seropositive for FeLV antigen. Fifty-eight cats (0.52%) were seropositive for both viruses. The probability of a positive test for FIV was not significantly higher for cats tested at shelters or rescue programs than at veterinary clinics. The probability of a positive test result for FeLV for cats tested at veterinary clinics was $1.7\times$ higher ($P = 0.023$) than for cats tested at shelters or rescue programs.

There were some statistically significant regional differences in seroprevalence for both FeLV and FIV. Seroprevalence for FeLV infection was significantly higher in Quebec (6.56%) than in British Columbia (2.23%, $P = 0.0236$) and Ontario (2.57%, $P = 0.0143$). Seroprevalence for FIV infection was significantly ($P = 0.0481$) higher in Quebec (9.32%) than in Nova Scotia (2.49%).

Several factors were significantly associated with risk of FeLV and FIV seropositivity. Adult cats were significantly more likely ($P < 0.001$) to be seropositive (4.43% FeLV, 5.86% FIV) than juvenile cats (1.69% FeLV, 1.56% FIV). Cats with current illness were significantly more likely ($P < 0.001$) to be seropositive (6.61% FeLV, 6.67% FIV) than healthy cats (1.99% FeLV, 3.22% FIV). The probability of a positive test result for FIV was 3.4 \times higher ($P < 0.001$) in cats with access to outdoors when compared to cats with no outdoor access. The probability of a positive test result for FeLV was 1.4 \times higher ($P = 0.027$) in cats with access to outdoors when compared with cats which had no outdoor access. The probability of a positive test result for FIV was highest in intact males (7.43%), and for FeLV was highest in intact females (7.29%) and intact males (7.08%). The probability of a positive test result for FIV or FeLV was 2.5 \times higher in samples seropositive for the other retrovirus than in seronegative samples ($P < 0.001$).

Discussion

The AAFP recommends that cats be tested for both FeLV and FIV infection under various circumstances, such as when first acquired, when exposed to a known retrovirus-infected cat, and before initial vaccination against FeLV or FIV (1). Sick cats should be tested even if previously tested negative. Cats at ongoing risk of infection should be tested annually.

To the authors' knowledge, this is the first broadly based study of retrovirus seroprevalence in Canada. It is not possible to compare the results from the current study with the previous studies of FeLV and FIV infection in Canada due to differences in the study populations, selection biases, test methodologies, and study design. In addition, while prevalence rates may change over time, trends should be monitored by repeating similar surveys.

The seroprevalence for both FeLV and FIV in this study was higher in cats with access to outdoors compared with cats which had no outdoor access, in intact cats compared with neutered cats, in adult cats compared with juvenile cats, and in sick cats compared with healthy cats. These findings are in accordance with similar North American retrovirus seroprevalence studies (3,7). Bite wounds due to territorial or sexual aggression are an efficient mode of retrovirus transmission. Bite wounds are more common in intact cats than neutered cats, as well as in cats with access to the outdoors. Cats with no access to outdoors are less likely to have contact with seropositive cats than are cats kept totally indoors. Neutered cats are also more likely to be kept indoors as companion animals and are at lower risk for retrovirus exposure. Both FeLV and FIV cause immunosuppression and are associated with many disease manifestations in cats (1), and are therefore expected to be more prevalent in sick than healthy cats.

The seroprevalence of FIV (4.3%) and FeLV (3.4%) in this study was higher than in a recent similar study of samples from predominantly US cats (3). In the US study, the overall prevalence of FIV was 2.5% (409/18 038) and FeLV was 2.3% (446/18 038). Samples from 325 Canadian cats were included in the study, with a prevalence of 3.1% for FIV and 2.5% for FeLV. The prevalence of co-infected cats was similar in both studies. The difference in seroprevalence between the studies may be influenced by the recruitment methods used for potential participants. In the US study, participants were recruited from a wider pool, including members of the AAFP and lists of animal shelters, cat rescue programs, and trap-neuter-return programs derived from Internet directories. In the current study, recruitment was focused on those centres already known to employ retrovirus testing as a matter of convenience for developing a list of potential participants that would cover private veterinary practices, humane organizations, animal shelters, and cat rescue programs.

The manufacturer reports point-of-care assay sensitivities for FeLV antigen and FIV antibody of 97.6% and 100% respectively, and specificities of 99.1% and 99.5% respectively (Package insert, SNAP Combo FeLV antigen/FIV antibody, IDEXX Laboratories). An independent study found 100% sensitivity and specificity for FIV antibody when unvaccinated cats were tested (8). Positive results for FeLV antigen obtained by ELISA testing should be confirmed with a secondary test such as an immunofluorescent antibody test or a polymerase chain reaction (PCR) assay and positive results for FIV antibody obtained by ELISA testing should be confirmed with a western blot assay (1). Investigation of the sensitivity and specificity of PCR assays for FIV offered by some laboratories has shown widely variable results (9,10), precluding recommendation of this test methodology. Positive test results were not confirmed using an alternate assay in the present study, so it is possible that false positive test results were included in the analysis. Feline immunodeficiency virus antibodies may be detected in uninfected cats that have been vaccinated against FIV (8) and in kittens with passively acquired immunity from an infected or vaccinated queen (11). An inactivated dual-subtype vaccine against FIV became available in 2003 in Canada (Fel-O-Vax FIV; Fort Dodge Animal Health, Overland Park, Kansas, USA), but FIV vaccination status was not recorded or known for cats in the present study. It is presumed that veterinarians would be unlikely to test cats for FIV if they were known to be vaccinated, and the population of cats tested by animal shelters/rescue organizations is unlikely to have a high rate of FIV vaccination. Therefore, it seems likely that bias of FIV prevalence estimates caused by vaccination was minimal. After exposure to FIV, seroconversion may take 60 d or longer (12), and similarly, after exposure to FeLV, seroconversion with detection by soluble antigen tests may take 30 d (13) so that some false negative results may also have been included for both viruses. False negative test results may also occur if the concentration of FeLV antigen or FIV antibody is below the detection limit of the test, or if the test is performed incorrectly.

Unadjusted seroprevalence data should be interpreted with caution because veterinarians and organizations may have

varying opinions about the importance of testing, and about criteria for selection of cats for testing despite the existence of testing guidelines. Although veterinarians may recommend testing for all cats, it is the individual cat owner who makes the final decision. It is reasonable to consider that sick cats are more likely to be tested than healthy cats as part of a diagnostic plan. Seroprevalence for both FeLV and FIV infection was higher in sick cats than healthy cats in the present study, similar to findings in other studies, but it is possible that these rates are artificially increased by selection bias.

Risk factor associations for FeLV and FIV seroprevalence were similar to those previously identified, but should also be interpreted with caution because cats and study participants were not selected in a random manner. For example, owned cats that never receive veterinary care or unowned cats that do not enter a shelter or come under the care of a rescue program were not sampled as part of this study. This limits the extent to which the findings can be generalized to the Canadian cat population. However, analysis identified age, gender, health status, and lifestyle as significant risk factors for FeLV and FIV seropositivity in agreement with other studies, including a recently published study of North American cats (3). This information can be used to counsel owners on prevention of disease transmission, such as by limiting access to outdoors and neutering.

Although testing for FeLV and FIV has been readily available for many years in Canada, and vaccines against FeLV have been in widespread use for more than 15 y, this study indicates that retroviral infections remain common in Canada. Increased awareness of Canadian seroprevalence data among veterinarians, animal shelters, rescue organizations, and pet owners may help improve testing and vaccination rates. Currently available guidelines for feline retrovirus testing and management developed by the AAFP should be adopted in Canada. CVJ

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