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Risk factors for seropositivity to feline retroviruses among owned domestic cats in Valdivia, southern Chile

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We identified risk factors associated with seropositivity to feline leukaemia virus (FeLV) or feline immunodeficiency virus (FIV) and the association between seropositivity to these retroviruses and the presence of clinical signs. Cats under veterinary care had lower risk of FeLV seropositivity and male cats had higher risk of FIV seropositivity. FeLV seropositive animals had higher odds of non-specific clinical signs and reproductive disorders. FIV seropositive cats had higher odds of buccal alterations. These findings are useful to obtain a first approach to identifying felines that need the application of diagnostic tests for retroviral infections.

Keywords: Feline leukaemia virus, feline immunodeficiency virus, immunochromatography, risk factors, seropositivity.

FELINE leukaemia virus (FeLV) and feline immunodeficiency virus (FIV) are important pathogens among domestic cat populations worldwide. Infections caused by these viruses are frequently present in clinical practice and they are associated with a high morbidity[1,2].

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Clinically, FeLV infection is related to a variety of syndromes, including neoplasia development, degenerative alterations of the hematopoietic cells and dysfunction of the immune system[3,4]. FIV infection affects both the humoral and cell-mediated immunity and consequently induces severe immunodeficiency, which leads to different clinical signs compared to FeLV infection[2,5].

Studies on populations of domestic cats provide an understanding of the frequency, distribution and characteristics of feline retroviral infections in different geographic locations[6]. Some reported prevalences for FeLV infection include 1.8% in Germany[7], 2.6% in USA[8] and 7.5% in Mexico[9]. For FIV infection some published prevalences include 11.6% in Italy[10], 19.5% in Turkey[11] and 31.3% in Malaysia[12].

In Chile, FeLV and FIV infections were detected in some surveys[13–16], but these studies did not determine risk factors associated with seropositivity. Thus, the aims of the present study were: (1) to identify possible risk factors associated with seropositivity for FeLV and FIV infection in a sample of owned cats from Valdivia, Chile, and (2) to determine the association between seropositivity for FeLV or FIV infection and the presence of clinical signs.

The study area was the city of Valdivia in Los Ríos region in southern Chile, with population of 140,559 habitants[13,17]. This city is located at 39°48′S and 73°14′W[18]. This research was the second part of a prevalence study on feline retroviruses aimed to estimate the prevalence of FeLV infection, FIV infection and coinfections, and determine risk factors associated with seropositivity for FeLV or FIV infection. The sample size was determined for a proportion (prevalence) using the Win EPI 2.0 software with the following data: an estimated population of 15,000 pet cats, based on the results of a feline census conducted in Valdivia (there is no information about feral cat population in the city)[19], an expected prevalence of 50% (ref. 20), an accepted error of 10% and a confidence level of 95% (ref. 21). The calculated sample size was 93 animals, but finally 124 cats were sampled considering a division of the city into nine neighbourhoods. The cat owners voluntarily agreed to participate in the study.

The methods for animal handling and blood extraction followed the guidelines for animal management and welfare of the Bioethics Committee at the Universidad Austral de Chile, Valdivia.

The diagnostic test used was the Speed DUO FeLV/FIV (Bio Veto Test laboratory, France). This is a lateral flow assay, based on immunochromatography that detects separately FeLV capsid antigen (p27) and antibodies directed against FIV transmembrane protein (gp40). The test was performed according to the manufacturer's recommendations.

For FeLV testing, 17/124 (13.7%) samples tested positive. For FIV testing 14/124 (11.3%) samples tested positive. Three samples (3/124; 2,45) tested positive for both FeLV and FIV. Due to the low seropositivity for coinfections, logistic regression analysis of this group was not performed.

All cat owners were interviewed by a veterinary practitioner using a structured questionnaire to obtain information about breed (breed versus mixed breed), gender (male versus female), age (young versus adult), reproductive status (neutered versus non-neutered), veterinary care (animals with veterinary care versus animals without veterinary care), vaccination status (feline herpesvirus 1, feline calicivirus, feline panleukopenia virus, rabies, FeLV), indoor versus outdoor habitat and number of cats at home (one versus more than one cat).

Requested additional information dealt with observations of the owners about behaviour and lifestyle of their cats: if the cat had gone outdoors without supervision (yes or no); if the cat had ever been lost (yes or no), if the cat lived or had lived with other cats of unknown FeLV and/or FIV status (yes or no), if it had fought with other cats of unknown FeLV and/or FIV status (yes or no), if the animal had been a stray cat (yes or no), and if the owner had adopted recently (in the last six months) a new cat with unknown FeLV and/or FIV status (yes or no).

All sampled cats were subjected to physical examination by a veterinary practitioner. The animals were categorized as having clinical signs suggestive of FeLV or FIV infection if they showed one or more of the following signs: non-specific signs (depression, anorexia or fever), gastrointestinal and/or abdominal alterations (vomiting, diarrhoea, abdominal masses, abdominal sensitivity or weight loss), respiratory system alterations (coughing, sneezing or nasal secretion), urinary system alterations (renal sensitivity, renomegaly, polyuria, polydipsia or disuria), reproductive disorders (infertility or abortions), ocular disease (conjunctivitis or ocular discharge), buccal alterations (halitosis/gingivitis, stomatitis or tooth loss), skin lesions (bite wounds or abscesses) and other clinical signs (e.g. lymphadenomegaly)[1,2,22,23].

Using Statistix 8, the association between the different characteristics of the cats (i.e. the cat owner interview results) and test results for FeLV or FIV was assessed using multivariate logistic regression modelling. For each infection, a separate model was constructed. The variables were first selected using univariate logistic regression for each variable ($P < 0.25$) and then, multivariate models were constructed using a forward strategy for variable inclusion. Goodness-of-fit of the models was assessed using the likelihood ratio test ($P < 0.05$).

The odds ratio (OR) and the 95% confidence intervals (95% CI) for the variables included in the multivariate models were estimated, considering statistically significant those variables that in the 95% CI did not include the non-effect level '1'.

The following interactions between variables on the basis of biological plausibility were assessed: (1) male cats + non-neutered cats; (2) male cats + if the animal had

gone outdoors without supervision (yes); (3) if the animal had gone outdoors without supervision (yes) + if the cat had been lost (yes); (4) non-neutered cats + if the cat had fought with other cats with unknown FeLV and/or FIV status (yes); (5) if it had been a stray cat (yes) + if it had fought with other cats with unknown FeLV and/or FIV status (yes), and (6) male cats + if the cat had been lost (yes).

Potential confounders were also assessed, considering bibliographic data and evaluation of the relative change in OR after inclusion of the potential confounders in the model. If this change was >10%, the variables were considered confounders[24].

Univariate logistic regression analyses were performed to assess the association between seropositivity to FeLV or FIV infection and the presence of clinical signs. OR and the 95% CI were calculated, assigning statistical significance to those variables that did not include the non-effect level '1' in the 95% CI.

For FeLV infection, the multivariate logistic regression model showed that being a cat under veterinary care was statistically associated with FeLV seronegativity (OR = 0.3) (Table 1). None of the evaluated interactions was statistically significant, and confounders were not found.

For FIV infection, the multivariate logistic regression model showed that being a male cat was statistically associated with the seropositivity. A non-neutered outdoor lifestyle and different neighbourhoods in the city of Valdivia, were found to be confounding variables (Table 1). None of the interacting variables showed statistical significance.

Out of the 124 sampled cats, 67 (54.0%) were classified as having clinical signs suggestive of FeLV or FIV infection. The univariate logistic regression analysis showed that seropositivity for FeLV was associated with the presence of 'non-specific clinical signs' (OR = 3.9) and with 'reproductive disorders' (OR = 22.7). For FIV infection, seropositivity was associated with the presence of 'buccal alterations' (OR = 4.3) (Table 2).

This study demonstrates that FeLV and FIV seropositivity are associated with different risk or protection factors. Being a cat with veterinary care was associated with a lower odds of seropositivity (OR = 0.3) compared to cats without veterinary care. The owners who provide veterinary care to their cats certainly show a greater interest in the health status of their pets. Conversely, a confounding variable included in the logistic regression model for FIV was the neighbourhoods in the city of Valdivia, because it is possible that the different socioeconomic status of the owners, associated with the neighbourhoods, could influence the health care and maintenance conditions of the cats and therefore, the probabilities of infection. Considering this, the information given by veterinary practitioners could help promote pet care, improving the management practices and increasing the probabilities of prevention, which could lead to a lower odds of infection. Some specific actions that can be taken to prevent FeLV and FIV infection are mentioned in the literature[5,25,26]. These are: (1) to promote the neutering of male and spaying of females to reduce aggressive behaviour; (2) indoor confinement of infected animals to reduce viral spread; (3) indoor confinement of FeLV and FIV uninfected pet cats to prevent exposure to infected stray and feral cats; (4) the application of diagnostic tests in individuals at risk to detect seropositive animals, and (5) vaccination.

Male cats were significantly more likely to be FIV-infected. This association has also been reported in studies in Italy[27], United Kingdom[28], Iran[29], Malaysia[12], Thailand[30], Australia[31], USA, Canada[32] and Australia[33]. Fighting over access to females for social rank establishment is common in male cats, increasing the probabilities of bite wounds and the transmission of FIV[34–37]. The aggressive behaviour involving territorial defence and a bold or proactive temperament could explain the risk of FIV infection[38,39].

Other confounding variables were also included in the multivariate logistic regression model for FIV. First, 'being an intact cat', a higher odd of seropositivity in sexually mature, non-neutered cats has been suggested in some feline retroviral studies[40,41]. The second variable was habitat 'outdoors', which can be explained because seropositivity to FIV could be higher in cats with outdoor access, according some authors[32,42].

In this study, the association between seropositivity to FeLV or FIV and the presence of clinical signs of disease was also evaluated. FeLV seropositive cats had 3.9 more odds to show unspecific clinical signs, such as depression, anorexia or fever, which are frequent findings in FeLV-infected cats[43].

FeLV infection was strongly associated with non-specific signs and reproductive disorders, consistent with previous findings[4,44]. Some queens develop an apparent infertility due to embryonic deaths or foetal resorption in the initial period of gestation, and abortions may occur later caused by endometritis or placentitis[43,45]. In persistently infected pregnant queens, FeLV could be transmitted to foetuses and kittens could also become infected after birth via colostrum or milk[26,46]. Some kittens could be immune to FeLV or conversely, they could be infected with the 'fading kitten syndrome', characterized by failure to nurse, dehydration, hypothermia and death at early age[1,44].

FIV infection was strongly associated with the presence of buccal alterations, which has been reported in surveys in Canada[47,48]. Gingivitis and periodontal disease may occur in up to 50% of FIV-infected cats and the presence of oral cavity disease increases the index of suspicion of FIV infection[49–51]. It has been suggested that buccal lesions could be a consequence of immunosuppression caused by FIV infection, which changes the

Table 1. Multivariate logistic regression models to identify risk factors associated with FeLV and FIV seropositivity

Variables	Categories	No. of sampled cats	OR (95% CI)
Model 1: FeLV			
Veterinary care	Without veterinary care	49	Reference
	With veterinary care	75	0.3 (0.1–0.9)[a]
Clinical signs	Without clinical signs	57	Reference
	With clinical signs	67	3.2 (0.9–10.6)
Model 2: FIV			
Sex	Female	67	Reference
	Male	57	4.7 (1.3–17.2)[a]
Reproductive status[b]	Neutered	67	Reference
	Non-neutered	57	0.3 (0.1–1.1)
Habitat at home: outdoors[b]	Yes	93	3.4 (0.4–25.6)
	No	31	Reference
Neighbourhood[b]	Sector 1	10	0.1 (0.0–3.9)
	Sector 2	8	0.2 (0.01–7.9)
	Sector 3	3	Reference
	Sector 4	19	0.1 (0.01–4.1)
	Sector 5	20	0.1 (0.01–3.2)
	Sector 6	18	0.2 (0.01–5.0)
	Sector 7	20	0.1 (0.01–4.1)
	Sector 8	15	0.3 (0.01–6.2)
	Sector 9	11	0.2 (0.01–6.9)

[a]Statistically significant; [b]Confounding variables.

Table 2. Univariate logistic regression analyses to determine associations between seropositivity to FeLV or FIV and presence of clinical signs

Clinical signs	Categories	No. of sampled cats	FeLV OR (95% CI)	FIV OR (95% CI)
Non-specific	Yes	28	3.9 (1.3–11.3)[a]	1.4 (0.4–5.0)
	No	96	Reference	Reference
Gastrointestinal and abdominal	Yes	15	2.7 (0.8–9.7)	0.5 (0.07–4.0)
	No	109	Reference	Reference
Respiratory system	Yes	17	1.4 (0.4–5.6)	0.5 (0.06–3.5)
	No	107	Reference	Reference
Urinary system	Yes	5	1.6 (0.2–15.0)	>999.9 (+∞)
	No	119	Reference	Reference
Reproductive	Yes	4	22.7 (2.2–232.2)[a]	>999.9 (+∞)
	No	120	Reference	Reference
Ocular	Yes	17	0.4 (0.04–2.9)	1.9 (0.5–7.5)
	No	107	Reference	Reference
Buccal	Yes	34	0.7 (0.2–2.6)	4.3 (1.4–13.6)[a]
	No	90	Reference	Reference
Skin	Yes	31	1.8 (0.6–5.3)	2.6 (0.8–8.0)
	No	93	Reference	Reference
Lymphadenomegaly	Yes	5	1.6 (0.2–15.0)	2.0 (0.2–19.4)
	No	119	Reference	Reference

[a]Statistically significant.

microflora in the buccal cavity and predisposes cats to infections with other pathogens such as feline calicivirus, possibly resulting in more severe oral cavity disease[52,53].

This study provides epidemiological information on FeLV and FIV infection in domestic cats in southern Chile. Being a cat under periodic veterinary care was a protective factor for FeLV infection and being a male cat was a risk factor for FIV infection. In addition, seropositivity to the viruses was related to different clinical signs, which emphasizes that regular evaluations of the health status of seropositive cats are essential. These findings could be useful to obtain a first approach to identifying suspected animals infected with FeLV or FIV in feline clinical practice, and/or for the application of diagnostic tests to detect retroviral infections.

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