

## The epidemiology features of retroviral infections in domestic cats from the Zagreb urban area

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### ABSTRACT

Feline immunodeficiency virus (FIV) and feline leukaemia virus (FeLV) are among the most common infectious diseases of cats and have a global impact on the health of domestic cats. Both viruses belong to the *Retroviridae* family and like other members of this family they are associated with lifelong infection after integration of the proviral DNA into the host cell genome. Prevalence data are necessary to define the risk factors, and prophylactic, management, diagnostic and therapeutic measures for stray and owned sick cats. In this study 324 domestic cats were tested with commercially available assays. The tested cats were divided into two groups, stray and owned sick cats. The overall percentage of seropositives for FIV infection was 18.51% and the prevalence for FeLV infection was 14.50%. FIV prevalence ranged from 13.13% in stray cats up to 20.88% within the sick owned cat group. The prevalence for FeLV infection was 6.06% in stray cats and 18.22% among sick owned cats. Regarding FIV infection, our study confirmed a significantly higher percentage of seropositives for male cats, as well as for sexually intact ones within the sick owned group. Males were significantly more likely to have positive results for both retroviral infections. The study confirmed the high rate of retroviral infections in cats from the Zagreb urban area. Males, sexually intact ones, and territorial aggression are predisposing factors for FIV infection, but not for FeLV. Preventive measures should include identification and segregation of infected cats, castration of outdoor male cats, and vaccination.

**Key words:** FIV; FeLV; prevalence; risk factors; prevention; Croatia

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## Introduction

Feline immunodeficiency virus (FIV) and feline leukemia virus (FeLV) are retroviruses causing two of the most important and common infectious diseases of cats. Both viruses are enveloped RNA viruses. FIV belongs to the *Lentivirus* genus, while FeLV is classified in the genus *Gammaretrovirus* (HOSIE et al., 2009; LUTZ et al., 2009; HARTMANN, 2011). As both viruses possess reverse transcriptase for integration of the provirus into the host cell genome, FIV and FeLV cause lethal, untreatable and lifelong infections. Luckily, retroviruses survive a short period of time outside the host and both viruses are readily inactivated by a variety of disinfectants (PEDERSEN, 1993; SYKES, 2014). Domestic and wild cats are affected hosts (TROYER et al., 2005; MELI et al., 2009). The major mode of FIV transmission is through biting wounds and to a lesser extent transplacental, through colostrum, blood transfusion and possibly venereal routes (YAMAMOTO, 1989; PARK et al., 1995; ROGERS and HOOVER, 1998; JORDAN et al., 1999). Transmission of FeLV primarily results from close contact with salivary secretions, such as through mutual grooming, licking and shared food and water dishes. Viremic cats may also shed the infectious virus in multiple body fluids, including nasal secretions, faeces, milk and urine. Evidence of horizontal transmission of FeLV by cat fleas also exist (HARDY et al., 1976; PACITTI et al., 1986; VOBIS et al., 2003; GOMES-KELLER et al., 2009). Although infected cats may experience a prolonged period of time with no clinical abnormalities, a variety of clinical and disease conditions are associated with retroviral infections, including anaemia, hematological disorders, lymphoma, and chronic inflammatory conditions, as well as susceptibility to secondary and opportunistic infections. Specific disease syndromes are associated with a very high rate, such as cutaneous abscesses (FeLV 8.8%, FIV 12.7%) and oral inflammation (FeLV 7.3%, FIV 7.9%) (BELLOWS and LACHTARA, 2006; GOLDKAMP et al., 2008; LEVY et al., 2008). Due to the nonspecific and various clinical manifestation as well as the possibly long asymptomatic stage of FIV and FeLV infection, every sick cat and those with unknown retroviral status should be tested (LEVY et al., 2008; LITTLE et al., 2011). Nowadays, plenty of different commercially assays are used, either enzyme-linked immunosorbent assay (ELISA) or based on an immunochromatographic principle, to reveal retroviral infection. Beside the commercially available assays, various polymerase chain reaction (PCR), Western immunoblotting, virus isolation and immunofluorescent antibody (IFA) assays, performed by experienced laboratories are used to detect retroviral status (LITTLE et al., 2011).

FIV and FeLV infections are endemic in the domestic cat population worldwide. The prevalence of FeLV infection has reportedly decreased over the past two decades, probably as a result of widespread testing programs, the development of commercially available diagnostic assays, and due to the implementation of effective vaccines (O'CONNOR et

al., 1991; MOORE et al., 2004; LEVY et al., 2006; STUTZER et al., 2011). On the other hand, the prevalence of FIV infection has not altered since the virus was discovered in 1986 (PEDERSEN et al., 1987). The vaccine against FIV was not introduced until 2002 and its efficiency is still under debate, while the heterogeneity of the gene sequences poses a great problem for developing a broadly affective and protective vaccine against different subtypes of FIV (ELYAR et al., 1997; LEVY et al., 2008; COLEMAN et al., 2014). Retroviral seroprevalence is highly variable in different geographical areas as well as in different domestic cat categories.

Estimated FIV prevalences vary from 1 to 14% in cats with no clinical signs, up to 44% in sick cats (HARTMANN, 1998). More precisely, the data indicate that the seroprevalence of FIV in Italy ranged between 24-30.9% in the sick cat population (BANDECCHI et al., 1992; PENNISI and BO, 1994), while 10.7-14.1% of clinically healthy cats were positive for FIV antibodies (PENNISI and BO, 1994; MAGI et al., 2002). Prevalence data obtained in Spain for FIV infection showed similar results, where 8.3% of healthy and 13.9% of clinically ill cats were positive (ARJONA et al., 2000). Other studies performed in Europe showed 27% FIV infected cats in Serbia (POTKONJAK et al., 2014), 33.3% in Slovenia (TOZON et al., 2008), 10.4% in England (MUIRDEN, 2002), and from 3.2% up to 8.4% in Germany (FUCHS et al., 1994; GLEICH et al., 2009). In Croatia, FIV was previously serologically confirmed in 3.3% of healthy and 10.3% of sick cats (KUČER et al., 2000).

For FeLV infection, the currently overall prevalence in the mixed cat population ranges from 1-6% (LEVY et al., 2006; GLEICH et al., 2009; LITTLE et al., 2009; HELLARD et al., 2011). A prevalence study for FeLV infection in Slovenia found 23.8% positive cats (TOZON et al., 2008), and a previous paper in Croatia showed 11.5% healthy and up to 14.9% clinically ill, positive cats (KUČER et al., 2000).

While FIV and FeLV can be life threatening viruses, proper management and treatment can give infected cats longer and healthier lives. The following study defines the current prevalence of FIV and FeLV in the Zagreb urban area, as well as determining predisposing factors and preventive measures for retroviral infections.

### **Materials and methods**

The research was performed on 324 domestic cats presented to the Department of Microbiology and Infectious Disease with Clinic at the Faculty of Veterinary Medicine, University of Zagreb. The tested cats were divided into two groups, stray (n = 99) and owned sick cats (n = 225). All stray cats have to be tested for retroviral infections prior to admittance to rescue shelters and afterwards to adoption programs. The majority of stray cats had no clinical abnormalities. Owned cats presented to the Clinic had health conditions which could be related to FIV and FeLV infection, so besides clinical treatment

all these cats were tested. Within the owned sick cat group there were 125 males (n = 32 castrated males; n = 93 sexually intact males) and 100 female tested cats. All tested cats included in this study were older than 6 months, therefore maternal antibodies (stemming from an infection or vaccination of the mother) could not interfere with true retroviral status. Peripheral blood samples were obtained from v. cephalica antebrachii or v. jugularis into EDTA-coated tubes by an aseptic method. To confirm retroviral infection, commercially available assays (FASTest® FIV; FASTest® FeLV, MEGACOR Diagnostic GmbH, Austria) were used to evaluate 324 blood samples. FASTest® FIV is a rapid immunochromatographic test for the qualitative detection of FIV antibodies in whole blood, plasma or serum of cats. Also FASTest® FeLV is a rapid immunochromatographic test for the qualitative detection of FeLV group specific antigens (p27) in the whole blood, plasma or serum of cats. The reported sensitivity and specificity for FASTest® FIV were 96.4% and 99.2%, and for FASTest® FeLV 94.7% and 98.8% (HARTMANN et al., 2007). Diagnostic tests were performed immediately after blood sampling according to the manufacturer's instructions. All data were summarized using percentages, while for statistical comparison and analysis the  $\chi^2$  test was used. A values of  $P < 0.01$ ,  $P < 0.05$  and  $P < 0.10$  were considered significant. Due to inconsistent data for stray cats regarding sex, neutered status and age, only 225 owned sick cats were used for statistical analyses to evaluate risk factors.

## Results

The overall (n = 324) percentage of seropositives seroprevalence for FIV was 18.51% (n = 60), and for FeLV 14.50% (n = 47). The percentage of seropositives for FIV infection ranged from 13.13% (n = 13) in stray cats up to 20.88% (n = 47) in the owned sick cat population. On the other hand, the prevalence for FeLV infection was 6.06% (n = 6) in stray cats and 18.22% (n = 41) in owned sick cats. The percentage of seropositives for FIV infection was significantly ( $P < 0.01$ ) higher in males (31.20%; n = 39) than in females (8.00%; n = 8) in the owned sick cats' group. Furthermore, we found a significant difference ( $P < 0.10$ ) in FIV infection between sexually intact (35.48%; n = 33) and neutered males (18.75%; n = 6) within the group of owned sick cats. The prevalence of FeLV infection in males (19.20%; n = 24) and females (17.00%; n = 17) was not significantly different ( $P = 0.67$ ) amongst the owned sick cats. In our study we did not find any significant difference ( $P = 0.26$ ) in FeLV infection between neutered (12.50%; n = 4) and sexually intact (21.50%; n = 20) males amongst the owned sick cats. Among the 225 tested owned sick cats 4.88% (n = 11) were positive for both retroviral infections. Males (8.00%; n = 10) were significantly ( $P < 0.05$ ) more likely to have positive results for both retroviral infections. The average ages of the owned sick cats were 7.9 years for FIV infected males and 6.1 years for females, while the average age for FeLV infection for both sexes was 4.3 years.

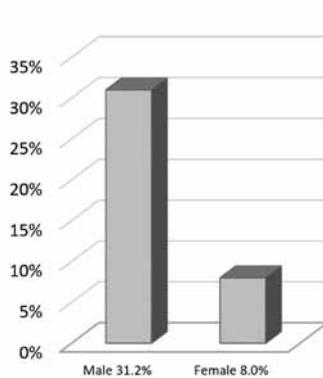


Fig. 1. Significantly higher seroprevalence in males for FIV infection

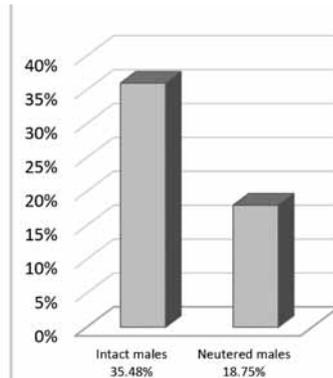


Fig. 2. Significantly higher seroprevalence in sexually intact males for FIV infection

Table 1. Prevalence of FIV and FeLV in the tested owned sick cats' group

Categories	Prevalence	
	FIV	FeLV
Sexually intact males	33/93 (35.48%) <sup>a</sup>	20/93 (21.50%)
Neutered males	6/32 (18.75%) <sup>a</sup>	4/32 (12.50%)
Total males	39/125 (31.20%) <sup>b</sup>	24/125 (19.20%)
Total females	8/100 (8.00%) <sup>b</sup>	17/100 (17.00%)
Total	47/225 (20.88%)	41/225 (18.22%)

<sup>a</sup> Significantly higher percentage of seropositives in sexually intact males in comparison with neutered males ( $P < 0.10$ ); <sup>b</sup> Significantly higher percentage of seropositives in males than in females ( $P < 0.01$ )

### Discussion and conclusions

Feline immunodeficiency virus and feline leukaemia virus have worldwide distribution among domestic cats. Both viruses can remarkably alter the cat's life. Due to the relatively long incubation period, the diversity of clinical manifestations and possible extended asymptomatic stage of the infection, FIV and FeLV are often left undiagnosed. Lately owner awareness about retroviral infections is increasing, but there are still reasons for poor compliance among veterinarians and cat owners, possibly due to their incomplete understanding of the lifelong nature of retrovirus infection, insufficient communication skills, the inconvenience of physical re-evaluation of cats, and finally the cost of testing (LITTLE et al., 2011). For these reasons every sick cat and those with unknown retroviral status should be tested.

Our study confirmed a high rate of retroviral infections in cats from the Zagreb urban area. The high FIV and FeLV prevalence is mainly a repercussion of the high density cat population in the urban area, the negligible number of vaccinated cats, the large number of stray cats, and generally infrequent testing.

This paper stresses the risk factors for FIV infection, which include male gender and outdoor access. Biting and territorial aggression are highly efficient modes for FIV transmission. Therefore, the natural territorial fighting behaviours of sexually intact male cats predispose them to a higher rate of FIV infection, which is confirmed in this research. This correlation between male sex and higher seroprevalence for FIV infection has been confirmed by several studies (ISHIDA et al., 1989; PERI et al., 1994; LEVY et al., 2006). The major mode for FeLV transmission is through saliva, milk, blood and urine, both vertically and horizontally. The difference between males and females regarding FeLV infection is usually not as large as for FIV (LEE et al., 2002). In our study, we did not find any significant difference for FeLV infection according to gender.

While testing and identification of infected cats are key tools for retroviral prevention of transmission, vaccination also play an important role. The combined use of testing and vaccination programs is assumed to have reduced the prevalence of FeLV infection over the past 20 years (O'CONNOR et al., 1991; MOORE et al., 2004; LEVY et al., 2006; LITTLE et al., 2011). Recommendations for use of the available FeLV vaccines have been published (RICHARDS et al., 2006; LEVY et al., 2008). Recommendations for FIV vaccines have also been published (RICHARDS et al., 2006; LEVY et al., 2008) but their efficiency is still under debate. Another important step for retroviral prevention is castration of all male cats that have outdoor access.

Further, more detailed research of retroviral prevalence and phylogenetic characterisation of viral strains are essential for developing better diagnostic tools, as well as highly effective prophylactic protocols.

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#### SAŽETAK

Infekcije virusom mačje imunodeficijencije i virusom mačje leukemije pripadaju među najčešće zarazne bolesti mačaka i znatno utječu na njihovo zdravlje. Oba virusa pripadaju porodici Retroviridae. Kao i ostali virusi iz iste porodice, uzrokuju perzistentnu infekciju ugradnjom provirusne DNK u genom domaćina. Podaci o proširenosti obiju infekcija nužni su za utvrđivanje čimbenika rizika zaraze, mjera prevencije te dijagnostičkih metoda i postupaka sa zaraženom životinjom. Za potrebe ovog istraživanja pretražena su ukupno 324 uzorka pune krvi mačaka komercijalno dostupnim dijagnostičkim testovima. Pretraživane mačke podijeljene su u dvije skupine, slobodnoživuće mačke i bolesne mačke koje imaju vlasnika. Od ukupnog broja pretraženih mačaka infekciju mačjim virusom imunodeficijencije dokazali smo u 18,51 % mačaka, dok je infekcija virusom mačje leukemije dokazana u 14,50 % pretraženih životinja. Proširenost infekcije mačjim virusom imunodeficijencije potvrđena je u 13,13 % mačaka unutar skupine slobodnoživućih mačaka, dok je 20,88 % bolesnih mačaka koje imaju vlasnika bilo pozitivno na istu zaraznu bolest. S druge strane, 6,06 % slobodnoživućih mačaka te 18,22 % bolesnih mačaka koje imaju vlasnike bilo je zaraženo virusom mačje leukemije. Istraživanje potvrđuje statistički značajnu proširenost virusa mačje imunodeficijencije u muških jedinki posebno onih nekastriranih. Muške jedinke također imaju statistički veću mogućnost zaraze obama retrovirusima. Provedeno istraživanje dokazuje visoku proširenost retrovirusnih infekcija među mačkama na području grada Zagreba. Utvrđeni čimbenici rizika zaraze virusom imunodeficijencije uključuju muški spol, nekastrirane muške jedinke te slododno držanje mačaka. Ti čimbenici rizika nisu dokazani za infekciju mačjim virusom leukemije. Mjere sprečavanja širenja obiju retrovirusnih bolesti uključuju testiranje mačaka, izdvajanje i izolaciju zaraženih, kastraciju muških mačaka te imunoprofilaksu izloženih jedinki.

**Ključne riječi:** virus mačje imunodeficijencije; virus mačje leukemije; proširenost; čimbenici rizika; prevencija; Hrvatska

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