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Transmission of feline immunodeficiency virus (FIV) among cohabiting cats in two cat rescue shelters

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ABSTRACT

Conflicting accounts have been published in the veterinary literature regarding transmission of feline immunodeficiency virus (FIV) between cohabiting cats in mixed households, and the mechanics of possible casual transmission, if it occurs, are poorly understood. Similarly, there are conflicting reports of vertical transmission of FIV. The aim of the present study was to document the FIV serological status of cats taken into two rescue shelters. At rescue shelter 1 (Rescue 1), cats cohabited in a multi-cat household of FIV-negative and naturally-infected, FIV-positive cats. A study was performed that combined a retrospective review of records of FIV serological status at intake (Test 1) and prospective FIV serological testing (Tests 2 and 3). Retrospective records were analyzed at rescue shelter 2 (Rescue 2), where FIV-positive queens with litters of nursing kittens were taken into the shelter, before being rehomed. FIV serology was performed on all kittens after weaning.

Initial test results (Test 1) for 138 cohabiting cats from Rescue 1 showed that there were 130 FIV-negative cats and eight FIV-positive cats (six male neutered and two female spayed). A second test (Test 2), performed in 45 of the FIV-negative and five of the FIV-positive cats at median 28 months after Test 1 (range, 1 month to 8.8 years) showed that results were unchanged. Similarly, a third test (Test 3), performed in four of the original FeLV-negative cats and one remaining FIV-positive cat at median 38 months after Test 1 (range, 4 months to 4 years), also showed that results were unchanged. These results show a lack of evidence of FIV transmission, despite years of exposure to naturally-infected, FIV-positive cats in a mixed household. At Rescue 2, records were available from five FIV-positive queens with 19 kittens. All 19 kittens tested FIV-negative, suggesting that vertical transmission had not occurred.

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Introduction

Feline immunodeficiency virus (FIV) is a retrovirus of the genus *Lentivirus*, which is endemic in cat populations worldwide. While the main mode of transmission is via bite wounds, vertical or perinatal transmission (in utero, intrapartum, milk/colostral) and transmission between cats in stable households, is much less common (Shelton et al., 1989). Under laboratory conditions, FIV is also infectious via parenteral routes, such as intravenous, intraperitoneal, intradermal and subcutaneous (Burkhard and Dean, 2003). Mucosal transmission of experimental infections has also resulted from virus inoculation into the nasal cavity, mouth, vagina and rectum, but transmission of FIV across mucosal surfaces is relatively ineffective compared to human immunodeficiency virus (HIV; Bishop et al., 1996). Previously published studies of closed 'mixed households' of cohabiting FIV-negative and FIV-positive cats have reported variable infection transmission rates (Table 1), but the prevalence

of episodes of inter-cat aggression was usually not quantified and the observation periods were often relatively short.

There have been a number of observational studies of vertical transmission of naturally-acquired FIV infections (Yamamoto et al., 1988; Ueland and Nesse, 1992; Medeiros et al., 2012) and experimental studies using FIV to develop models of vertical transmission of HIV (Yamamoto et al., 1988; Sellon et al., 1994; O'Neil et al., 1996; Rogers and Hoover, 1998). The study by Ueland and Nesse (1992) was unable to demonstrate vertical or horizontal transmission of FIV among 25 adult cats (6 FIV-positive, 19 FIV-negative) and 48 kittens (30 of which were born to FIV-positive queens) in a closed breeding colony over a 9 month period. A later study, using molecular methods and serology, demonstrated vertical transmission of naturally-acquired infection from one queen to one kitten (Medeiros et al., 2012). Chronically-infected queens in one experimental model transmitted the virus to approximately half of their offspring via late in utero, intrapartum and milk-borne routes (O'Neil et al., 1996), and approximately 60% of full term fetuses were infected in another chronic infection model (Rogers and Hoover, 1998). Milk-borne infection was demonstrated at a similar rate (62.5%) in kittens born to acutely-infected queens (Sellon et al., 1994). Another study dem-

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Table 1
Published studies of closed 'mixed' populations of FIV-negative and FIV-positive cats.

Reference	FIV-negative (n)	Originally FIV-positive (n)	Originally FIV-negative cats that became infected (n)	Laboratory/home	Observation period
Yamamoto et al., 1988	14	18	0	Laboratory	4–14 months
Shelton et al., 1989	31	16	0	Home	Median of 2 years
Shelton et al., 1990	68	5	0	Home	3.5 years
Dandekar et al., 1992	20	NR	1 ^a	Laboratory	2–4 years
Addie et al., 2000	17	9	6	Home	10 years

NR, not recorded.

^a In this study, 10/19 cats that remained serologically FIV-negative at the end of the study period tested FIV-positive using PCR.

onstrated that maternally-derived anti-FIV antibodies from either vaccinated or infected queens could prevent infection in neonatal kittens inoculated intraperitoneally (Pu et al., 1995).

The aims of the present study were to investigate horizontal transmission of naturally-acquired FIV between cats in a mixed, multi-cat household and to investigate viral transmission from naturally-infected FIV-positive queens to their kittens.

Materials and methods

Study design

Information was collected from two cat-only rescue shelters (designated Rescue 1 and Rescue 2). At Rescue 1, a retrospective review of records of FIV serological status at intake (Test 1, $n = 138$) was performed for all cats that had entered the shelter since its inception in October 2003. Some cats had also received a second and a third FIV serological test (Test 2, $n = 16$; Test 3, $n = 2$). Prospective FIV serological testing was performed on all cats that were still present at the shelter on 16 November 2011 (Test 2, $n = 34$; Test 3, $n = 3$). Additional information was also obtained regarding housing status (indoor-only vs. access to outdoors) and any history of inter-cat aggression for the duration of each cat's stay at Rescue 1. At Rescue 2, records were reviewed for information relating to FIV-positive pregnant and nursing queens and their kittens that had been presented to the shelter.

Animals and FIV testing

Approval for this study was granted by the Purdue University Animal Care and Use Committee (Protocol number 1201000568; 9 February, 2010).

For Rescue 1, cats cohabited in a single household, sharing litter pans, food/water dishes and bedding. This was a privately owned rescue facility based in a domestic home, run by single caregiver. Cats were sourced from local partner shelters after they were spayed/neutered and they left the rescue facility when adopted. All cats were housed indoors only, except for one cat (Cat A; FIV-positive at Test 1), which was allowed occasional outdoor access. None of the cats were known to have any history of vaccination against FIV. Records were available from initial serological testing (Test 1) for feline leukemia virus (FeLV) and FIV (SNAP FIV/FeLV Combo Test, IDEXX Laboratories) from 138 cats at the rescue facility. All 138 cats in the study were FIV tested on the day they entered the shelter, except for one cat that was determined to be FIV-positive 17 months before shelter entry and four FIV-negative cats that tested FIV-negative at 4 months ($n = 2$), 11 months ($n = 1$) and 18 months ($n = 1$) before shelter entry. These cats were still FIV-negative at 14 months ($n = 2$) and 2 months ($n = 2$) after shelter entry, respectively. Veterinarians and staff at the shelters where the cats were originally sourced performed Test 1 at the time of spay/neuter, before the cats arrived at the rescue.

After adoptions from Rescue 1, 50 of the original 138 cats that underwent Test 1 still remained at the rescue facility and these underwent a second test (Test 2; IDEXX SNAP FIV/FeLV Combo Test) a median of 28 months after Test 1 (range, 1 month to 8 years 10 months; interquartile range 15 months to 3 years 9 months). The rescue facility caregiver performed Test 2 on 13/50 cats and a certified veterinary technician tested the remaining 37 cats. PCR testing for FIV (RealPCR Test for FIV, IDEXX Laboratories) was also performed on 33/50 cats, using the same blood collected for Test 2. Personnel performing the follow-up tests were aware of previous test results.

A third test (Test 3; IDEXX FIV/FeLV SNAP Combo Test) was performed on 5/50 cats that underwent Test 2. These five cats had been continuously housed at the rescue facility since entry and initial testing. Test 3 was undertaken a median of 3 months after Test 2 (range, 1–3 years 9 months; interquartile range 1 month – 2 years 6 months). The same veterinary technician who performed Test 2 performed Test 3.

Rescue 2 was a privately-owned, feline-only rescue facility, which accepted stray and owner-relinquished cats. Cats were assessed by a resident veterinarian and serological testing (IDEXX FIV/FeLV SNAP Combo Test) was performed on entry. Any heavily pregnant FIV-positive queens were transferred to foster homes, once initial assessment and testing had been completed. While in foster care, queens and their litters did not have access to any other FIV-positive cats. Queens were returned to the rescue facility for ovariectomy after their kittens had been weaned at ap-

proximately 8 weeks of age. Kittens were tested for FIV/FeLV (IDEXX FIV/FeLV SNAP Combo Test) after weaning. Queens and kittens then offered for adoption if deemed healthy by the rescue veterinarian. Vaccination against FIV was not performed at the rescue center, or while the cats were in foster care.

Data analysis

Month-by-month infection pressure was calculated by tracking the number of FIV-positive and FIV-negative cats cohabiting each month since the first FIV-positive cat entered the shelter (March 2008). Calculations for FIV-negative cats that were adopted during the study period assumed that the adoption date (rather than the most recent test date) was the end date of the calculation, so that complete residence time in the population was used for the calculation. Descriptive statistics were calculated using commercially available software (GraphPad Prism 5 for Mac OS X).

Results

Rescue 1

Information on cats recruited into the study and results of FIV testing are presented in Table 2. For each cat tested, the FIV serological status remained the same at all time-points assessed. It was reported that one of the FIV-positive cats (Cat A; male neutered [MN] domestic shorthair [DSH], aged approximately 2 years at Test 1; residing in the household for 2 years and 4 months during the study period) was frequently aggressive toward other cats, but skin wounds were not noted on either cat after episodes of aggression. Another two FIV-positive cats (Cat B, MN DSH, aged approximately 2.5 years at Test 1, resided in the household for 1 year and 4 months during the study period; Cat C, female spayed [FS] DSH, aged 5 months at Test 1, resided in the household for 3 years and 8 months during study period) habitually groomed other cohabiting cats.

At Test 1 ($n = 138$) and Test 3 ($n = 5$), all cats tested FeLV-negative. At Test 2 ($n = 50$), one cat (Cat D; MN DSH, aged approximately 8 years at Test 2) tested FeLV-positive and FIV-negative. Cat D was

Table 2

Descriptive information for cats tested for FIV/FeLV (IDEXX FIV/FeLV SNAP Combo Test) at Rescue 1.

	FIV-negative cats	FIV-positive cats
Test 1 (n)	130	8
Test 1 – Median age ^a	4 months	28 months
(range)	(2 months – 13 years)	(5 months – 11 years)
Test 1 – Gender ^b	71 MN, 59 FS	6 MN, 2 FS
Test 2 (n) ^c	45	5
Test 2 – Median age ^a	4 months	23 months
(range)	(2 months – 8 years)	(5 – 31 months)
Test 2 – Gender ^b	25 MN, 20 FS	4 MN, 1 FS
Test 3 – n ^c	4	1
Test 3 – Median age ^a	30 months	4 years
(range)	(7 months – 4 years)	
Test 3 – Gender ^b	2 MN, 2 FS	1 FS

MN, male neuter; FS, female spayed.

^a The veterinarians and staff at the source shelters from which the cats were obtained estimated cat age at the time of spay/neuter.

^b Gender refers to gender status at the time of admission to the private rescue facility.

^c Cats tested at Test 2 were a subset of cats tested at Test 1. Cats tested at Test 3 were a subset of cats tested at Test 2.

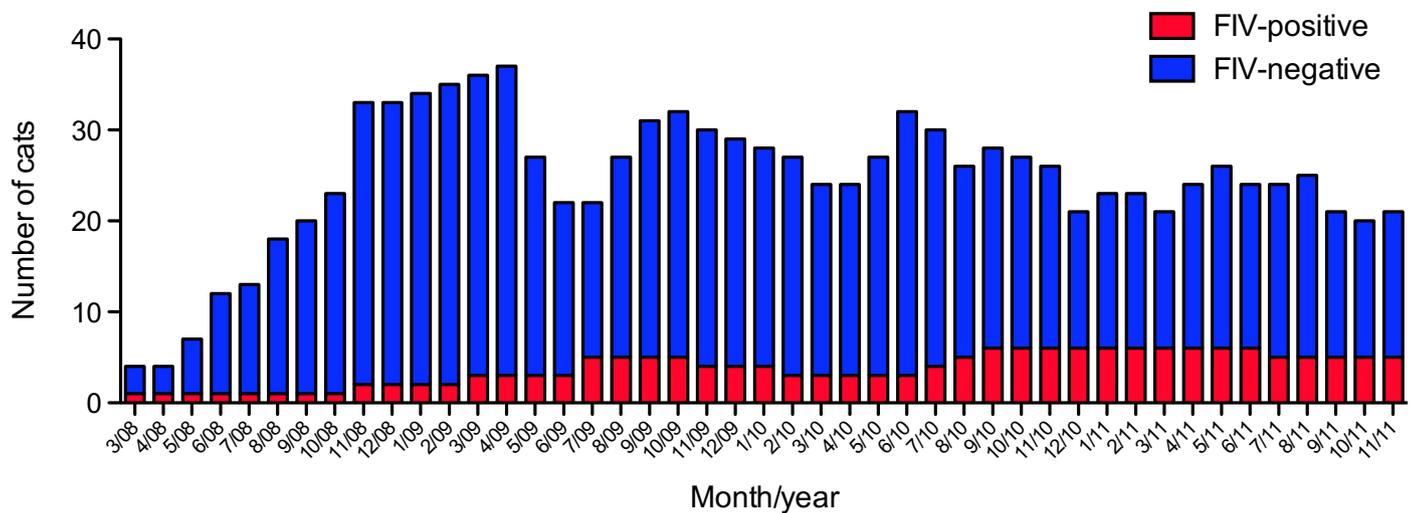


Fig. 1. Number of FIV-positive and FIV-negative cats cohabiting at Rescue 1 month by month (March 2008–November 2011).

the only cat that tested FeLV-positive at Rescue 1 during the study period. This cat had left the household after Test 1 was performed and was subsequently retested (Test 2) before being considered for readmission to the household, but because of the FeLV-positive result, the cat was not readmitted and died 2 months later.

Results of FIV testing by PCR, performed on whole blood samples collected for Test 2 in 33/50 cats showed that both ELISA and PCR results were negative in 29/33 cats; both ELISA and PCR results were positive in 3/33 cats, and the remaining cat tested ELISA-positive but PCR-negative.

Infection pressure results are presented in Figs. 1 and 2. The ratio of FIV-negative to FIV-positive cats cohabiting at Rescue 1 month by month (March 2008–November 2011) ranged from 2.5–22.0, while the total number of cats ranged from 4 to 37 over the study period.

Rescue 2

Serological test results from five queens and their 19 kittens from Rescue 2 are presented in Table 3. All five queens were presented as strays, so their previous vaccination history was unknown.

Although demonstrated to be FIV positive by serology, FIV PCR or other FIV confirmatory testing was not performed.

Discussion

In this study of cats cohabiting in a mixed household over a period of months to years, despite mutual grooming, mild aggression, shared food bowls, litter boxes, bedding etc. there was no evidence of transmission of infection from FIV-positive to FIV-negative cats. Additionally, at a second rescue shelter, serological testing did not demonstrate any evidence of vertical transmission of FIV from naturally-infected mothers to their kittens. The number of infectious units transmitted (Kusuhara et al., 2005) and the viral phenotype (Burkhard and Dean, 2003) are likely to be important factors that determine the risk of infection following exposure, but while relative virulence has been investigated for experimental strains (Dean et al., 1999; Pedersen et al., 2001), this information is not known for field strains of FIV causing naturally-acquired infections.

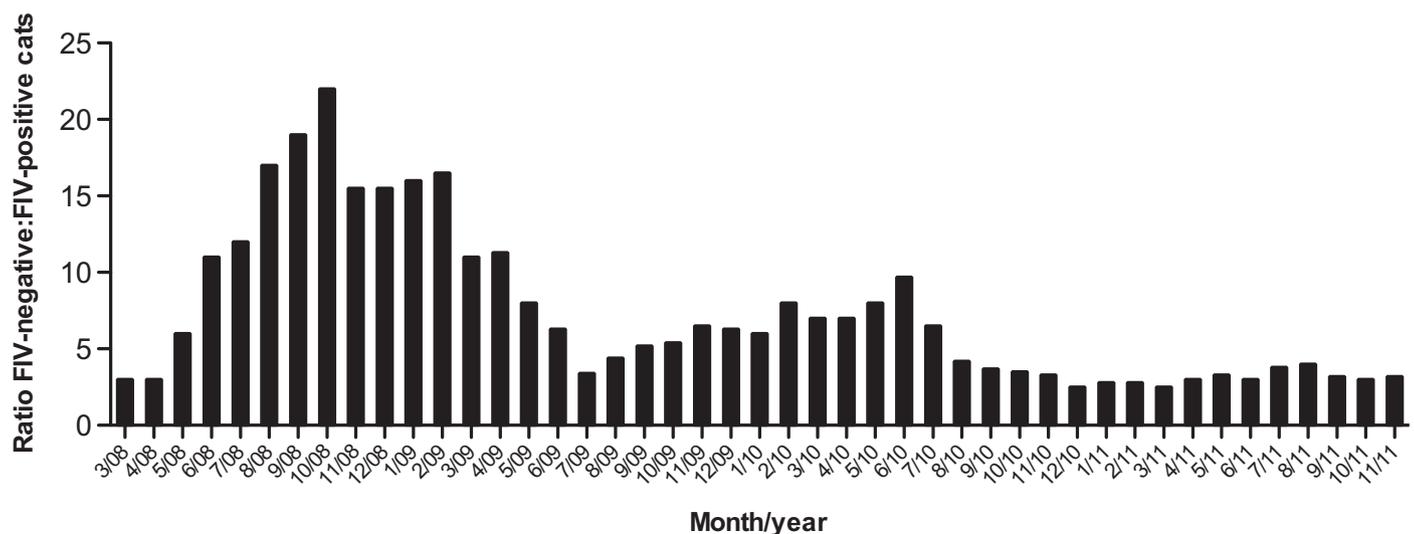


Fig. 2. Ratio of FIV-negative to FIV-positive cats cohabiting at Rescue 1 month by month (March 2008–November 2011).

Table 3
Details of serological testing (IDEXX FIV/FeLV SNAP Combo Test) of FIV-positive queens and their weaned kittens from Rescue 2.

	Litter size and genders	Age of kittens at first test (weeks)	Test result	Age of kittens at second test (weeks)	Test result
Cat A	n = 5, 3 females, 2 males	9	All kittens negative	NP	NP
Cat B	n = 3, 2 females, 1 male	13	All kittens negative	26	All kittens negative
Cat C	n = 4, 3 females, 1 male	25	All kittens negative	NP	NP
Cat D	n = 3, 1 female, 2 males	17	All kittens negative	NP	NP
Cat E	n = 4, 2 females, 2 males	13	All kittens negative	NP	NP

NP, not performed.

Another important risk factor for FIV transmission in a multi-cat household is the frequency of aggression events, resulting in bite wounds (Shelton et al., 1989). All of the cats at Rescue 1 were spayed or neutered and kept indoors (except for one FIV-positive cat), thereby reducing the risk of territorial aggression; the caregiver at Rescue 1 reported no penetrating bite wounds in any of the cohabiting cats. This is supported by the results of a large prevalence study that reported sexually intact status and outdoor lifestyle as major risk factors for FIV infection (Levy et al., 2006) and other studies, which have linked cat bite wounds and abscesses with FIV-positive status (Goldkamp et al., 2008; Chang-Fung-Martel et al., 2013). Additionally, it is of note that the median age of FIV-negative cats from Rescue 1 at Test 1 was 4 months, since kittens are known to be a low risk group for FIV (Levy et al., 2006), probably because territorial aggression has not yet developed. Therefore, recommendations for 'mixed' housing of FIV-positive and FIV-negative cats should be grounded in the important considerations of feline behavior, neuter status, virology and immunology.

Because FIV is transmitted directly between cats, the ratio of cohabiting FIV-positive to FIV-negative cats is an important factor in the risk for disease transmission. Additionally, if cats are stressed, because there are large numbers kept together with few opportunities to escape from one another, the risk of fighting is likely to increase. At Rescue 1, the ratio and the total number of cats varied widely over the study period, but FIV transmission was not demonstrated in the population.

ELISA-negative, PCR-positive FIV infection was not demonstrated in the 33 cats tested using both methods at Rescue 1. Dandekar et al. (1992) reported that in some FIV-negative cats cohabiting with FIV-positive cats, FIV amplicons were detectable by PCR (Table 1). Those individuals did not develop a serological response or clinical signs of disease, although other studies have failed to substantiate this phenomenon. It is possible that delayed seroconversion could explain our results, although all but 3/29 FIV-negative cats that were tested by PCR had been cohabiting with FIV-positive cats for at least 10 months when molecular analysis was performed. Alternatively, the cats that tested FIV-negative by PCR could have had undetectable amounts of viral nucleic acid in their blood, thus making their FIV status difficult to identify using PCR.

In the present study, vertical transmission of FIV was not evident. However, since further confirmatory testing was not performed at Rescue 2, it is possible that some of the FIV-positive ELISA test results might be due to an antibody response following FIV vaccination, rather than via natural infection. However, since all these cats presented as intact strays, this was considered unlikely. This is supported by another recent study, which reported that <40% of cats had antibody titers against viral agents present in core vaccines for cats (feline panleukopenia virus, feline calicivirus and feline herpesvirus) at the time of admission to rescue shelters and factors associated with seropositivity included being neutered and owner relinquished (DiGangi et al., 2012). It is also interesting to note that vertical transmission seem to be much less common in published reports of cats with naturally-acquired FIV (Ueland and Nesse, 1992;

Medeiros et al., 2012) than for experimental infections (Sellon et al., 1994; O'Neil et al., 1996; Rogers and Hoover, 1998).

This study has a number of limitations. Firstly, there were only a small proportion of cats for which longitudinal FIV serological data were available from the original 138 cats tested at Rescue 1 (Test 2 n = 50/138; Test 3 n = 5/138). This was due to a combination of an active adoption program at that shelter and the long period of time over which FIV test records were collected (since the shelter opened in October 2003 until the study was performed in November 2011). Additionally, personnel performing the tests were not blinded to previous test results, although it seems unlikely that this could influence their interpretation of the results with this type of test (Levy et al., 2004; Hartmann et al., 2007). Since all available cats were tested, knowledge of previous test results did not impact on selection of cats for testing. Finally, it is possible that misclassification error occurred if cats testing FIV-negative had been recently infected but seroconversion had not occurred by the time of FIV testing.

Conclusions

FIV serological status in cats cohabiting in a mixed household of FIV-negative and naturally infected FIV-positive cats at Rescue 1 did not change over a period of years, despite unrestricted access to one another, mutual grooming, minor episodes of aggression and sharing food and water dishes, litter pans and bedding. Serological results did not demonstrate any evidence of vertical transmission of FIV from five FIV-positive queens to their 19 weaned kittens at Rescue 2. These study findings could have implications for the recommendations made by veterinarians and shelter staff asked to advise cat owners or adopters contemplating co-housing FIV-positive and FIV-negative cats and to shelters caring for litters of kittens born to FIV-positive queens. However, careful management is required when cats are first introduced to one another, as the potential for agonistic interactions that could result in FIV transmission is increased. Because of this, it is important to determine FIV status before cats are introduced to one another and then to observe interactions until the likelihood of aggression resulting in penetrating bite wounds is considered negligible. If there is a reasonable suspicion that such agonistic interactions will occur when cats are left unsupervised, FIV-positive and FIV-negative cats should be segregated from one another.

Conflict of interest statement

The author of this paper has no financial or personal relationship with other people or organizations that could inappropriately influence or bias the content of the paper.

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