INFLUENCE OF INFECTIOUS PRESSURE ON THE PATHOGENESIS OF FELINE LEUKEMIA VIRUS INFECTION

University of Zurich

University of Bern | University of Zürich
vetsuisse-faculty

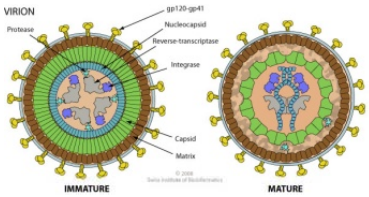
A. Major[1], V. Cattori[1], E. Boenzli[1], P. Ossent[2], M. Meli[1], R. Hofmann-Lehmann[1], H. Lutz[1]

[1]Clinical Laboratory, Vetsuisse Faculty, University of Zurich, Switzerland
[2]Institute for Veterinary Pathology, Vetsuisse Faculty, University of Zurich, Switzerland

Introduction

FeLV is a gammaretrovirus naturally occurring worldwide in domestic cats and some related small felids. FeLV transmission occurs mainly via the oronasal route. The virus first replicates in tonsils and local lymph nodes. Infected lymphocytes spread the virus to the bone marrow. It is generally believed that infection of the bone marrow is compulsory for infection to become systemic [1].

Following transmission, different outcomes have been observed [2-4].



Gomes-Keller et al. at our laboratory demonstrated [5] that naive cats exposed to feces of persistently infected cats, seroconverted, indicating infection, but remained negative for the presence of FeLV provirus and p27 in blood. FeLV DNA sequences were found in some tissues.

This is an outcome of FeLV infection so far not described.

It appears that low infectious pressure may lead to infection without detectable involvement of the bone marrow.

Aim

This study aimed to further characterize the course of infection after exposure to low infectious pressure, and to test the hypothesis that, depending on infectious dose, there may exist a so far unknown pathogenesis of infection without involvement of the bone marrow.

Materials & Methods

Animals. *3 groups of 7 SPF cats each were once infected oronasally with 1'000, 10'000 and 100'000 FFUs of FeLV A (Glasgow-1 strain), respectively. 5 SPF cats were kept as negative controls.*

Sample collection. *EDTA blood was collected weekly until week 6 post-infection (p.i.), later biweekly until week 20 p.i..*

FeLV viral RNA loads in plasma and proviral loads in blood & tissues. *Extraction by MagNa Pure, quantification by Taqman real-time RT-PCR and PCR, respectively [6].*

FeLV p27 antigen load. *p27 was measured by Sandwich ELISA [7].*

Antibody assays. *Anti-FeLV p45 and anti-FeLV whole virus antibodies were detected by indirect sandwich ELISA [3,8]. Seroconversion was assumed if OD values during the last 6 weeks p.i. of the observation period were outside the 95% tolerance interval for the control cats.*

Virus isolation from bone marrow and tissues *was performed in vitro. Supernatants of bone marrow cells and inoculated FEA cells respectively were tested by p27 ELISA [5].*

Lymphocyte proliferation assay. *PBMCs were stimulated with virus, concanavalinA, or cell culture medium. IFN-γ transcription was assessed by real-time RT-PCR and ELISPOT assay [9].*

Whole blood stimulation. *In week 18 p.i., heparinized blood was stimulated with virus, and IL-2 and fIFN- γ transcription was determined by real-time RT-PCR[10].*

Results

ELISA for detection of FeLV p27 in plasma

Only one cat was positive for FeLV p27 antigen. This cat that became transiently positive, developed persistent PCR-positivity (orange line, Fig 1).

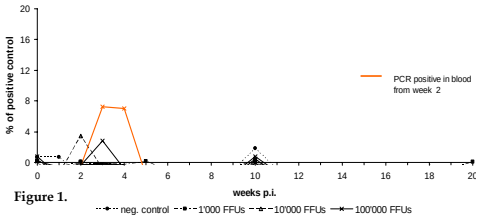


Figure 1.

Antibody assays

Two of 26 cats seroconverted using the whole virus ELISA (orange and green line), but not using the p45 ELISA.

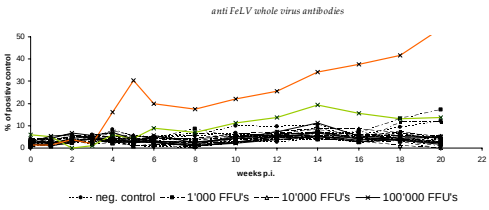


Figure 2. *PCR positive in blood from week 2*

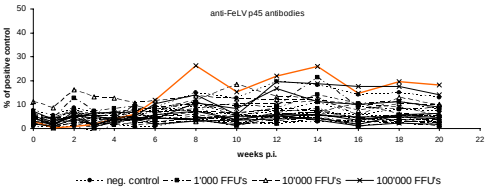


Figure 3. *PCR positiv in blood from week 2.*

Lymphocyte proliferation assay and whole blood stimulation

The feline IFnγ-ELISpot and the real-time PCR analysis of cellular response were negative for all cats tested.

Detection of proviral DNA in blood and viral RNA in plasma

Proviral DNA was detected in one of the cats (JCR2) that received 100'000 FFUs from week 2 p.i.. All other cats remained consistently PCR negative in blood.

Plasma samples of week 2, 3, and 4 p.i. were tested for viral RNA. Viral RNA was detected only in samples of the provirus-positive cat.

Detection of FeLV DNA sequences & virus isolation from tissues

FeLV DNA sequences were only found in the organs of the cat (JCR2) positive for FeLV provirus in blood (Fig. 4). All other organs were negative for provirus.

All cats tested negative for the presence of infectious virus in tissue samples of urinary bladder, myocardium, lungs, mesenterial lymph node, thymus, and bone marrow.

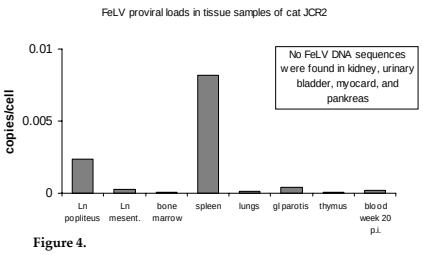


Figure 4.

Discussion

In this study we attempted to characterize the course of FeLV infection after exposure to low doses of FeLV.

In these experiments we observed one cat that became infected as concluded from persistent provirus-positivity and seroconversion. Another cat seroconverted without being detectably positive for provirus. All other cats did not show signs of infection.

In the study of Gomes-Keller et al., the cats were exposed to FeLV in feces continually over 26 weeks. In the present study, exposure to low dose challenge was once. It was concluded that a short and low dose exposure can result in an infection without involvement of the bone marrow. However, this type of infection may be rare.

Summary

Low infectious pressure of FeLV may result in p27- and PCR-negativity together with seroconversion.

Seroconversion may serve as marker for FeLV infection.

Acknowledgements

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