Retrovirus serology in snow leopards and other wild felids in European zoos

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In the domestic cat retroviruses are important pathogens and are known to occur at varying prevalence worldwide. The feline leukemia virus (FeLV) was isolated in 1964 (Jarrett et al. 1964), the feline spumavirus (also designated feline syncytium forming virus, FeSFV) was first described in 1969 (Riggs et al. 1969), and the feline immunodeficiency virus (FIV) was recently discovered by Pedersen et al. (1987). The disease spectrum of retroviruses in the domestic cat is very broad, including anemia, immune suppression and neoplasia caused by FeLV, immune suppression, gingivitis and a number of other clinical symptoms caused by FIV and gingivitis and possibly arthritis caused by FeSFV. After it was evident that FIV is widespread in the domestic cat we became interested in the question whether wild felids would also be infected by retroviruses. To this end we tested serum samples collected from wild cats including snow leopards in different zoos in Europe and one in India. We tested these samples for presence of antibodies to the three domestic cat retroviruses, and for antibodies to the equine infectious anemia virus (EIAV). EIAV is a lentivirus of horses inducing fever episodes at intervals of several months, leading to erythrocyte destruction and anemia. Although it is antigenically related, it is clearly distinguished from FIV by other criteria. In this study we found that some retrovirus infections are highly prevalent in wild felids. Because retrovirus infections tend to be latent for a prolonged time period there is always the potential danger that animals moved between zoos or even back to the wild habitat may spread a virus to the new environment where it formerly was not present. It is the goal of this report to highlight some of these aspects.

MATERIAL AND METHODS

Serum samples: a total of 124 serum samples collected from 12 species of wild cats were obtained from the following 10 zoos:

Royal Zoological Society of Antwerp, Belgium
Kolmardens Djurpark, Copenhagen, Denmark
Zoological Garden, Mulhouse, France
Allwetterzoo, Münster, Germany
Ruhr Zoo, Gelsenkirchen, Germany
Tierpark Hagenbeck, Hamburg, Germany
Zoologischer Garten, Basel, Switzerland
Zoo Mauerhofer, Frauenfeld, Switzerland
Zoologischer Garten, Zürich, Switzerland
Sakkarbaug Zoo, Junagadh, India

The 12 Asiatic lions in the Sakkarbaug Zoo were originally derived from the nearby Gir Forest Sanctuary. The samples from these lions were collected directly at Sakkarbaug Zoo. Serologic tests: FIV, FeLV and FeSFVantigens were produced in cell culture and purified by gradient centrifugation as described earlier (Lutz et al. 1988). Gradient purified EIAV was generously provided by Dr. S. Aaronson, National Cancer Institute, Bethesda, Maryland, USA. The sera were tested by ELISA using 200 ng of the purified antigens per test, by Western Blot assays (200 ng per nitrocellulose strip), and by IFA (FeSFVantibodies). A commercial conjugate (rabbit anticat IgG conjugated to horseradish peroxidase) was used in these assays. In addition, the sera were tested for presence of FeLV p27 antigen (Lutz et al. 1983).

RESULTS

Antibodies to FIV were readily detected in African but not in Asiatic lions (Table 1, Figure 1). In addition, antibodies to FIV were also found in one tiger and one jaguar but not in five snow leopards, all from the Zoo of Zürich. In some of the lion sera the antibody titers to FIV were very high and reached titers of higher than 1000 in Western Blots. The antibody pattern in Western Blots of lion sera was different from that of domestic cats. The lion antibodies recognized predominantly the core protein p24 and only to a small degree p17. None of the sera tested positive for FeLVantigen. Only two cheetahs were

positive for FeLVantibodies and these animals were previously vaccinated with a commercial FeLVvaccine. FeLV obviously does not play an important role in wild felids. Antibodies to FeSFV were found in low prevalence in tigers, cheetahs, panthers, lynxes and serval. One African lion was weakly positive for FeSFV. Antibodies to EIAV were found in all species tested with the exception of the lynxes (Table 1, Figure 2). Some, but not all of the EIAVpositive sera were positive for FIV. Six of 8 Indian lions that were tested for EIAVantibodies showed a reaction with EIAVgagproteins but not with FIVcomponents.

DISCUSSION

In our study we found that antibodies to FIV are highly prevalent in African lions kept in European zoos. In other wild felid species FIVantibodies were either not observed or only at a very low prevalence. It was concluded that the putative "lionimmunodeficiencyvirus" is antigenically closely related but clearly distinct from FIV. The arguments for this conclusion are: (1) the lionFIV could not be experimentally transmitted to domestic SPFcats (Lutz et al., 1992), (2) the lion sera showed a Western Blot pattern different from that of the domestic cat and (3) lion sera do not react with the envelope component of FIV (Letcher and O'Conner 1991) nor do they show virus neutralizing activity (Dr. M. Bendinelli, Pisa, pers. comm., 1992). From the observation that sera from Indian lions had antibodies to EIAV but not to

TABLE 1. Prevalence of retrovirus antibodies in wild felids.

Genus		Antibodies found to the following viruses FIV	FeLV	FeSFV	EIAV	
African lion (*Panthera leo*)	(53)	30/53 (57%)	0/53	1*/52	11/49 (27%)	
Indian lion (*Panthera leo goojratensis*)	(12)	0/12 (0%)		0/53	n.t.	6/8 (75%)
Tiger (*Panthera tigris*)	(20)	1/20 (5%)		0/20	4/19	5/20 (25%)
Cheetah (*Cinonynx jubatus*)	(13)	0/13	2**/13	4/13	8/13 (62%)	
Snow leopard (*Uncia uncia*)	(5)	0/5	0/5	0/5	1/5	
Leopard (*Panthera pardus*)	(6)	0/6	0/6	0/6	2/2	
Jaguar (*Panthera onca*)		(4)	1/4	0/4	1/4	4/4
Lynx (*Lynx lynx*)	(3)	0/3	0/3	1/3	0/2	
Ocelot (*Leopardus pardalis*)	(2)	0/2	0/1	0/1	1/1	
Puma (*Felis concolor*)		(2)	0/2	0/1	0/1	1/1
Serval (*Leptailurus serval*)	(3)	0/3	0/3	3/3	1/3	
Fossa (*Fanaloka fossa*)		(1)	0/1	0/1	0/1	1/1
Total		124				

* very weak result; ** the 2 positive animals had been vaccinated with Leukocell
n.t. not tested

FIV it may be concluded that an additional lentivirus may be of importance to wild cats that, up to now, has not been discovered.

Of special interest is the question from where the lentivirus of wild felids originates. Two reports from South Africa by J. Spencer (pers. comm., 1992) have shown that the lions in the Krüger National Park are FIVpositive (12/12 lions tested) whereas 66/66 lions tested in the Etosha National Park are FIVnegative. FIVinfection has also been found in the Serengeti National Park in Tansania (Dr. S. O'Brien, National Cancer Institute, pers. comm.). So it seems that the lion lentivirus originates from some clearly

defined areas in Africa. Letcher and O'Conner (1991) have shown that 16/22 Indian lions in Lincoln Park Zoo are FIVpositive. This is in contrast to our present data. The different findings can be explained either by the fact that the Asian lions of the Lincoln Park Zoo became infected by having contact with African lions in the zoo or that these lions are hybrids between African and Indian lions as suggested by O'Brien et al. (1987). Barr et al. (1989) found two of 17 snow leopards in Cheyenne Mountain Zoo, Colorado, to be infected with FIV. It is unclear whether these snow leopards became infected by close contact with African lions or whether snow leopards have their own "snow leopard lentivirus".

From the data discussed above it becomes clear that a number of questions regarding the breeding program of snow leopards and the different viruses, especially the FIVrelated lentivirus, have to be addressed: (1) How is the FIVrelated lentivirus spread among zookept wild felids? If it can be transmitted by droplets, this would

FIGURE 1. Western Blots with FIV (500 ng per strip) and various wild felid sera. Lane 1, SPF cat negative control serum; lane 2, FIV positive control serum. Note the strong anti-p24 and the weak anti-p17 reaction in lanes 21 and 22.

FIGURE 2. Western Blots with EIAV (500 ng per strip) and various wild felid sera. Lane 1, conjugate control; lane 2 positive horse serum; lane 3, negative cat control serum.

pose a serious problem to zoos in general. (2) What is the pathogenic potential of this lentivirus in wild felids? Could it be that the lion lentivirus is not a pathogen for lions and only becomes pathogenic when it crosses the species barrier? (3) Do snow leopards in the wild have their own lentivirus? (4) What measures should be taken to control the spread of retro and other virus infections when exchanging snow leopards between zoos?

This last question is relatively simple to answer. Prudence suggests that snow leopards should be tested for FIV and possibly spumavirus infections before they are brought into contact with other snow leopards. By eliminating FIV positive animals from breeding it should be possible to keep snow leopards (and other felids) free of FIV infection. It would be highly desirable that information regarding infectious diseases be recorded in the stud book. To help reach the goal of keeping snow leopards negative for FIV and other infections, the authors offer to test by Western Blot for FIV antibodies any snow leopard serum sent to their laboratory.

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