RESERVOIRS OF FELINE HAEMOPLASMAS

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Introduction

Three haemotropic mycoplasmas (aka haemoplasmas) have been identified in pet cats, designated *Mycoplasma haemofelis* (Mhf), '*Candidatus* Mycoplasma haemominutum' (CMhm) and '*Candidatus* Mycoplasma turicensis' (CMt). The latter was discovered in a Swiss pet cat and phylogenetic analysis revealed its close relationship to rodent haemoplasmas, which brought up the hypothesis of an interspecies transmission between mice and cats. Blood-sucking arthropods are suspected to be involved in the transmission of haemoplasmas between cats (1,2), but a recent experimental transmission between cats by fleas has not been conclusive (3). The aims of the present study were to investigate wild felids, ticks, fleas and mice as possible reservoirs of feline haemoplasmas and the molecular characterization of different haemoplasma isolates based on the 16S rRNA gene.

Materials & Methods

DNA was extracted from blood samples or serosanguinous fluid derived from 257 wild felids (Table 1), 192 free-ranging Swiss mice and from 73 cat fleas (*Ctenocephalides felis)*, 4 dog fleas (*Ctenocephalides canis)* and 86 *Ixodes* ticks collected from Swiss pet cats (Figure 3). The 110 Brazilian wild felids included 12 wild cat species kept in the São Paolo zoo and 1 free-ranging ocelot. All samples were analysed with real-time PCR assays for Mhf, CMhm and CMt infections (4). The 16S rRNA gene from 21 haemoplasma isolates from wild felids was sequenced and phylogenetic tree was constructed with the neighbour-joining method. Risk factors for hemoplasma infection were analyzed with Chi² or Fisher's Exact Test for categorical variables (origin) and with Kruskal-Wallis 1-way ANOVA by Ranks and the Dunn's post test for multiple comparisons for continuous variables (PCV, only available for Serengeti lions, Iberian lynx and Brazilian wild felids).

Table 1. Country and origin of the wild felids in this study.

Variable	Iberian lynx	Eurasian lynx	European wildcat	Serengeti lion	Brazilian wild felids
Country	Spain	Switzerland	France	Tanzania	Brazil
Zoo-kept Free-ranging	20/35 (57%) 15/35 (43%)	0/36 (0%) 36/36 (100%)	0/31 (0%) 31/31 (100%)	0/45 (0%) 45/45 (100%)	109/110 (99%) 1/110 (1%)

Results

Among the 257 wild felids, 37% tested PCR-positive for feline haemoplasma infection (Tables 2 and 3). All three feline haemoplasmas were detected in each sample group (European wildcat, Eurasian lynx, Iberian lynx, Serengeti lions and Brazilian wild felids), and co-infections were common. Six wild felid species tested negative for feline haemoplasmas, including 2 cheetahs (*Acinonyx jubatus*), 23 Jaguarundis (*Herpailurus yaguaroundi*), 4 leopards (*Panthera pardus*), 5 Pampas cats (*Oncifelis colocolo*), 11 Siberian tigers (*Panthera tigris altaica*) and 2 snow leopards (*Uncia uncia*). In phylogenetic analyses, most haemoplasma isolates showed close evolutionary relationship to pet cat derived isolates (Figure 1).

Free-ranging wild felids were significantly more often infected with feline haemoplasmas than zoo-kept animals (Figure 2). PCV values were not significantly different between haemoplasma uninfected animals and cats singly or co-infected with feline haemoplasmas.

Two out of 73 cat fleas and 3 out of 110 hard ticks tested PCR-positive for CMhm. No feline haemoplasmas were detected in the murine samples and in the dog fleas.

Table 2. Number and percentage of European wildcats, Eurasian lynx, Iberian lynx and Serengeti lions that tested real-time PCR-positive for Mhf, CMhm and CMt.

Species	Mhf positive (%)	CMhm positive (%)	CMt positive (%)
European wildcat *Felis silvestris silvestris*	1/31 (3)	6/31 (19)	11/31 (35)
Eurasian lynx *Lynx lynx*	4/36 (11)	14/36 (39)	2/36 (6)
Iberian lynx *Lynx pardinus*	7/35 (20)	9/35 (26)	3/35 (9)
Serengeti lion *Panthera leo*	31/45 (69)	43/45 (96)	34/45 (76)

Table 3. Number of different Brazilian wild felid species that tested haemoplasma real-time PCR-positive.

Species	Mhf positive	CMhm positive	CMt positive
Puma *Puma concolor*	0/2	1/2	0/2
Oncilla *Leopardus tigrinus*	0/33	3/33	0/33
Margay *Leopardus wiedii*	1/9	1/9	0/9
Geoffrey's cat *Oncifelis geoffroyi*	0/7	1/7	0/7
Lion *Panthera leo*	0/5	1/5	0/5
Ocelot *Leopardus pardalis*	1/7	4/7	1/7

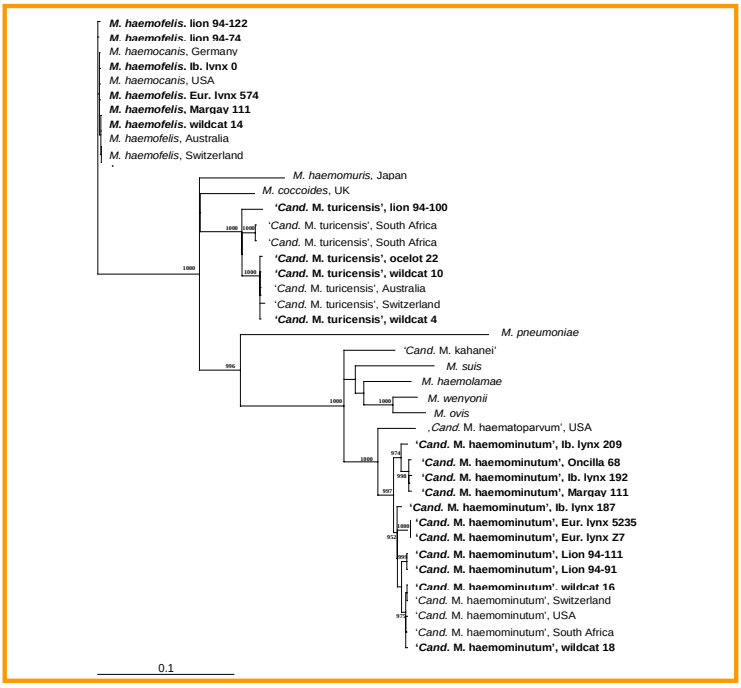

Figure 1. Phylogenetic analysis of the 16S rRNA gene sequences of Mhf, CMhm and CMt isolates from different wild felid species. Bootstrap values are given at the nodes of the tree; only values of ≥ 900 are shown.

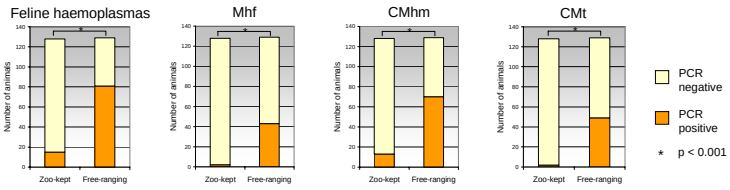

Figure 2. Significant association of feline haemoplasma infection with non-captive state of the animals.

Figure 3. Blood sucking arthropods as possible reservoirs of feline haemoplasmas.

Conclusions

Nine out of 15 wild felid species from three different continents were found to be infected with feline haemoplasmas. In contrast to an earlier study on 54 captive non-domestic cats (5), we found a remarkable high prevalence for these agents. The higher prevalence in free-ranging wild felids compared to zoo-kept animals could be explained by their more frequent exposure to arthropods and more common fight activity resulting in a higher infection risk with these agents (4, 6). Haemoplasma PCR-positive wild felids did not exhibit lower PCV values when compared to PCR-negative animals, but some of the animals showed anaemia. The lack of clear clinical signs despite of haemoplasma infections could be due to a chronic carrier state in these animals. However, a conclusion about the pathogenic potential of these agents in wild felids can not yet be drawn. In fleas and ticks, feline haemoplasmas were found at a very low frequency and our data so far do not confirm the hypothesis of an interspecies transmission of haemoplasmas between mice and cats.

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