Primary study of seroprevalence to virus pathogens in wild felids of South Primorie, Russia


Abstract: Seroprevalence to nine different virus pathogens was estimated for Russian big cats (Amur tiger (Panthera tigris altaica Temminck, 1844) and far-eastern leopard (Panthera pardus orientalis (Schlegel, 1857))) in Southern Primorie, Russia (n = 25), in 2008–2016. Serum samples from smaller cats (Eurasian lynx (Lynx lynx (Linnaeus, 1758)) and far-eastern wildcat (Prionailurus bengalensis euptilurus (Elliot, 1871))) were also tested for these pathogens (n = 19) during the same period. Felids of Russian Southern Primorie showed seroprevalence to eight out of nine tested pathogens, including highly dangerous feline immunodeficiency virus, feline leukemia virus, and canine distemper virus. Antibodies to feline panleukopenia virus were found to be much more widespread in cats (45%) than antibodies to any other virus. They were detected in samples taken from tigers, leopards, and the far-eastern wildcats but not lynxes. Antibodies to pseudorabies virus were detected only in Amur tiger (29%), whose main prey is the most common carrier of the virus (wild boar), unlike for the other studied cats’ species.

Key words: Amur tiger, Panthera tigris altaica, far-eastern leopard, Panthera pardus orientalis, Eurasian lynx, Lynx lynx, far-eastern wildcat, Prionailurus bengalensis euptilurus, seroprevalence, pathogens, canine distemper virus.

Résumé : La séroprévalence de neuf pathogènes viraux distincts a été estimée pour les grands félins de Russie (n = 25), dont le tigre de Sibérie (Panthera tigris altaica Temminck, 1844) et la panthère de Chine (Panthera pardus orientalis (Schlegel, 1857)), dans le sud du Primorî (Russie), de 2008 à 2016. Des échantillons sériques de félins plus petits (lynx boréal (Lynx lynx (Linnaeus, 1758)) et chat-léopard de Chine (Prionailurus bengalensis euptilurus (Elliot, 1871))) ont également été analysés pour ces pathogènes (n = 19) durant la même période. Les félidés du sud du Primorî russe présentent une séroprévalence pour huit des neuf pathogènes analysés, dont le très dangereux virus de l’immunodéficience feline, le virus de la leucose feline et le virus de la maladie de Carré. Des anticorps contre le virus de la panleucopénie féline se sont avérés beaucoup plus répandus chez les félins (45 %) que les anticorps contre tout autre virus. Ils ont été découvrez dans des échantillons prélevés de tigres, de panthères et de chats-léopards, mais pas de lynx. Des anticorps contre le virus de la pseudorabie ont été détectés seulement chez des tigres de Sibérie (29 %), dont la proie principale, le sanglier, est le vecteur le plus courant du virus, contrairement aux autres espèces de félins étudiées.


Introduction

Infectious diseases are a serious danger for wild animals that results in population decline (Roelke-Parker et al. 1996; Murray et al. 1999; Deem et al. 2001). However, little is known about the presence of infectious pathogens in the populations of carnivores in the Russian Far East, namely far-eastern leopard (Panthera pardus orientalis (Schlegel, 1857)), far-eastern wildcat (leopard cat) (Prionailurus bengalensis euptilurus (Elliot, 1871)), and Amur tiger (Panthera tigris altaica Temminck, 1844). Studies of seroprevalence in Amur tigers are sparse (Quigley et al. 2010; Goodrich et al. 2012a) and studies of far-eastern leopards are unique (Goodrich et al. 2012b). There are no data on the other two cats that inhabit the same area of Russia (far-eastern wildcat and Eurasian lynx (Lynx lynx (Linnaeus, 1758))). Seroprevalence to different pathogens in Amur tigers was studied only in the central part of the Russian Far East (Sikhote-Alin Reserve) (Goodrich et al. 2012a, 2012b), to five virus pathogens: feline panleukopenia virus (FPV), canine distemper virus (CDV), feline immunodeficiency virus (FIV), feline leukemia virus (FeLV), and feline coronavirus (FCoV). FPV affects rapidly proliferated cells resulting in diarrhea in domestic cats (Felis catus Linnaeus, 1758), suppression of marrow activity, and a decrease in immunity (Parrish 1995). FCoV is widely distributed in feral cats’ populations and contributes to the advancement of infectious peritonitis, which may be fatal for these animals (Pedersen 1987). CDV led to a population crash of lions (Panthera leo Linnaeus, 1758) in Serengeti and resulted in the deaths of 1000 individuals in 1994 (Roelke-Parker et al. 1996). FIV (homologous to the human immunodeficiency virus) was detected in do-
mestic cats and some wild cats (Brown et al. 1994) and has been proven to result in intense immunodepression due to a decrease in the number of T-lymphocytes (Torten et al. 1991). FeLV is a widely geographically distributed virus that leads to anemia, immunosuppression, and different forms of lymphoma (Hoover et al. 1975; Hardy et al. 1976). So far, the mortality of FeLV-infected cats has been described for Lynx, Puma, and Felis genera (Hoover and Mullins 1991; Sleeman et al. 2001; Cunningham et al. 2008).

Besides these pathogens, several other viruses pose a significant threat to the cats’ family. These are feline herpesvirus (FHV), feline calicivirus (FCV), influenza A virus, and Aujeszky’s disease virus (pseudorabies virus). FHV develops intensively in the case of concurrent infections and possible immunodeficiency (manul (Felis manul Pallas, 1776) (Ketz-Riley et al. 2003)) and may result in the death of some of felids (tiger) (Sun et al. 2014). It is widespread in feral cats’ populations and is transferred through amicable contact between animals (Hellard et al. 2013). In 2013, a young tiger female (about 5 months old) died in the Centre of Rehabilitation of tigers and other rare species in Alekseevka, Russian Far East, and FCV was proved to be the cause of death (M. Alsinetskii, Moscow Zoo Head Veterinarian, personal observation). Influenza A virus (variations of which include bird and porcine flu) may be transferred and spread by carnivores (Kuiken et al. 2004; Butler 2006) and could pose a danger to wild cats (Keawcharoen et al. 2004). The pseudorabies virus often leads to the death of carnivores (Lyubashenko et al. 1958; Ohshima et al. 1976), and the wild boar (Sus scrofa Linnaeus, 1758), the main carrier of this virus in the wild (Lari et al. 2006; Vengust et al. 2006; Wilson et al. 2009), is the main prey of tigers in the Russian Far East (Yudakov and Nikolaev 2012).

The distribution of these viruses is dependent on their ways of transmission and their lethality for the hosts and carriers. The main information about these viruses is compiled in Table 1. Some viruses are only spread among felids (FeLV, FCV, FIV, etc.) and some others are transmitted by prey species or other carnivores (CDV and Aujeszky disease virus) or a wide variety of other species (influenza A virus). The distribution of pathogens also depends on survival of the viruses in the environment.

Seroprevalence may show interspecific differences that relate to host ecology and sensitivity to these viruses (Pavlova et al. 2015, 2016). Four different wild cat species reside in the Russian Far East, all of them listed in the IUCN Red List: far-eastern leopard as critically endangered, Amur tiger as endangered, Eurasian lynx as least concern (game species in Russia), and far-eastern wildcat as least concern (at the species level, for Bengal leopard cat (for the subspecies (far-eastern wildcat), no evaluation was made). Three species (Amur tiger, Eurasian lynx, and far-eastern wildcat) inhabit all of the Southern Primorie, and the far-eastern leopard inhabits only the southwest of Primorie (National Park “Land of the Leopard”). The total number of far-eastern leopards has increased over the last 10 years (about 70 individuals in Russia, 57 in the National Park “Land of the Leopard” (Virtkalova and Shevtsova 2016)); a snow survey showed that the Amur tiger population is currently comprised of approximately 500 animals in Russia, with roughly 40 of them inhabiting the studied area. The numbers of Eurasian lynx and far-eastern wildcats are unknown but camera trapping shows that both of these species are even less widespread than tigers. Three large cats (including lynx) prey mainly on different ungulate species (Nippon deer (Cervus nippon Temminck, 1838) and Siberian roe deer (Capreolus capreolus (Pallas, 1771))), tigers and leopards also hunt wild boars, and tigers in the Ussuriskii Reserve prey upon the red deer (Cervus elaphus Linnaeus, 1758). All of these cats also prey on small carnivores (Asiatic badger (Meles leucurus (Hodgson, 1847)) and raccoon dog (Nyctereutes procyonoides (Gray, 1834))) (Pikunov and Korkishko 1992; Yudakov and Nikolaev 2012). Tigers may also prey on bears (Ursus arctos Linnaeus 1758 and Ursus thibetanus G. [Baron] Cuvier, 1823) and occasionally kill leopards and lynxes (Yudakov and Nikolaev 2012). Lynxes prey mainly on small-sized ungulates, and when their density is low, lynxes prey on lagomorphs and birds (Matsushkin 1978). As opposed to large cats, the far-eastern wildcat mainly hunts rodents, amphibians, and reptiles and inhabits mixed forests with open meadows near streams and small ponds (Yudin 2015). The other three species live in the mixed and deciduous forests, usually avoiding open areas. Consequently, all four species live in the same area, in adjacent habitats, hunt similar species of prey (at least three of them), and encounter each other or excretes of other cat species (and, in rare cases, even prey on other cat species). These factors all benefit cross-species virus transmission. All species may come into con-

<table>
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<td>Feline herpes virus</td>
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tact with domestic cats and dogs (tigers and far-eastern wildcats come closer to the villages during winter, but feral cats and dogs are also seen regularly inside the forest).

Thus, four cat species exist in similar habitats but differ in feeding ecology. Early study of Amur tiger diseases at the middle part of the range showed the presence of antibodies to five different viruses (Goodrich et al. 2012a). The aim of this study was to estimate the occurrence of antibodies to the same (and four more) viruses in tiger and three other cat species and compare them on the basis of the ecology of these animals with the ultimate goal of understanding the transmission of viruses in this region. We suggested that all four cat species may show similar seroprevalence to the same pathogens because of the similar habitats of these species. Inter-specific differences could be explained by the differences in feeding ecology or species-specific sensitivity to the viruses.

**Materials and methods**

This study was conducted at the south of Primorski Krai (Russia) in the Ussuriskii Reserve of the Russian Academy of Sciences (2008–2016) and National Park “The Land of Leopard” (2010–2016) (Fig. 1). Average altitude of this area is 300–400 m above sea level (rarely, up to 600 m). The study covered a total of approximately 1500 km². This area is characterized by low winter temperatures (average temperature in January is −17.9 °C) and warm summers (August temperature is +19.7 °C) with high humidity levels (70%–80%). The objects of study were four species of Felidae: Amur tiger (n = 17 (10 males, one of them 16–17 months old, and 7 females), far-eastern leopard (n = 8 (four males and four females, two of the females 1–1.5 years old)), Eurasian lynx (n = 4 (three males and one female)), and far-eastern wildcat (n = 15 (12 males and three females)). All animals were adults except the ones mentioned specifically. Tigers, leopards, and lynxes were captured using Aldrich foot snares (Rozhnov et al. 2013) that were set for tiger and leopard trapping. One lynx and all far-eastern wildcats were captured in box-traps (120 cm x 80 cm x 80 cm) using bait (meat or fish). Tigers and leopards were immobilized with injector DAN-INJECT SP25 (Dan-Inject, Denmark) with 0.1% medetomidine (“Domitor", Orion,
Sulikhan et al. (2018). This showed us very high (tiger) or growing test systems of “ImmunoComb” (Biogal, Galed Labs. Acs Ltd, Kibbutz Ga’al, Israel) with consecutive computer estimation of the antibody titer (animals with a titer of 1:16 for FHV, 1:20 for FCoV, 1:32 for FCV, and 1:80 for FPV were considered positive) (Soft Immunocomb, Israel). According to the manufacturer’s information, sensitivity and specificity of these kits for domestic cats were 84% and 95% for FHV, 96% and 94% for FCoV, 96% and 89% for FCV, and 90% and 98% for FPV. We also tested these kits (for FPV, FHV, and FCV) on vaccinated and nonvaccinated lynxes, wildcats, and tigers (at Tcchernogolovka Station (Naidenko 2005) and in the Centre of Rehabilitation of tigers and other rare species (Aleksvekvk) for 5–12 individuals of each species, showing positive results only for previously vaccinated species. Seroprevalence to influenza virus A and pseudorabies virus was estimated with ELIA (“cut-off” method) using commercial kits (Vetbiochim (Narvak), Moscow, Russia) (sensitivity of the kits was 97.6% and 98.2%, respectively, and specificity was 99.6% and 99.8%, respectively). In this case, protein NP of influenza A virus and glycoprotein gB of pseudorabies virus were immobilized at the wells’ bottom. The method of competitive ELISA was used with the monoclonal antibodies (to NP and gB) conjugated with horseradish peroxidase and serum antibodies. These kits were never tested specifically for cats but they were developed to detect antibodies to pathogens in a wide variety of mammalian species. Testing of these kits on breeding colonies of domestic cats, far-eastern wildcats, and lynxes at the Tcchernogolovka Station (Naidenko 2005) did not show any individuals seropositive to these pathogens. The presence of FeLV antigens and FIV antibodies was detected with the immunochromatography method using “speed tests” (BVT, France) (Naidenko et al. 2014; Pavlova et al. 2015). According to the manufacturer, this test’s sensitivity and specificity are both higher than 94% (sensitivity: FeLV = 94.7%, FIV = 96.3%; specificity: FeLV = 99.2%, FIV = 98.9%). It is necessary to note that seropositivity (antibody presence) to different pathogens does not guarantee that the animal is afflicted or even a carrier of the pathogen, only that the individual came into contact with this pathogen at some point (like with CDV). In some other cases, the presence of antibodies may reflect current infections (like with FIV). Moreover, in some cases, antibodies are produced to the pathogen that is not virulent for the host species (in our case, most likely, CDV for far-eastern wildcat).

Statistical analysis was conducted using Cochran’s Q test for related samples to compare the level of serum prevalence to different viruses in the same species. To compare the percentage of seropositive animals between species, the two-tailed Fisher’s exact test was used. The 95% level of confidence was calculated and p < 0.05 was considered to be statistically significant. Statistical analyses were conducted using Microsoft Excel (Microsoft Corporation, Redmond, Washington, USA) and Statistica version 8.0 (StatSoft, Inc., Tulsa, Oklahoma, USA).

### Results

In the course of this study, the antibodies to eight of nine tested viruses dangerous for cats were detected in felids in South Primorie (Table 2). There were no animals seropositive to FCoV. Antibodies against eight other viruses were found in one to three feline
species. We found that serum prevalence varied significantly for different viruses (Cochran’s Q test: $Q_{b}=55.60, p < 0.001$) among all cat samples. This fact could be explained by the differences in seroprevalence to different viruses in tigers ($Q_{b}=41.32, p < 0.001$). In leopards and far-eastern wildcats, these differences were not significant (correspondingly $Q_{b}=14.43, p = 0.072$ and $Q_{b}=12.00, p < 0.151$). Among all samples, seroprevalence of FPV was much higher than that of any other virus (Fisher’s exact test: $p = 0.000$–$0.021$). Seroprevalence to FCoV was also higher than seroprevalence to FCoV, FIV, FeLV, FHV, and influenza A virus (Fisher’s exact test: $p = 0.001$–$0.034$). No other differences in seroprevalence to viruses were found among all cat samples.

FPV is likely the most widespread virus out of the ones we tested for. Eleven serum samples from Amur tigers (six males and five females) and three samples (one male and two females) of leopards were positive to FPV. Six far-eastern wildcats were positive to FPV as well. There were no animals positive to FPV in lynxes, which differs significantly from the results in tigers (Fisher’s exact test: $p = 0.035$). Among all cats, 45% of serum samples were positive to FPV ($n = 44$).

Three serum samples of Amur tiger males and one sample of a far-eastern leopard male were seropositive to FCoV. Six far-eastern wildcats were seropositive to FCoV ($n = 15$), this statistic being only marginally higher than among tigers (Fisher’s exact test: $p < 0.127$). Total percentage of seropositive samples was 23% ($n = 44$).

Antibodies to pseudorabies were detected in five samples of Amur tigers (four males and one female) and were not found in any other species (Fisher’s exact test: $p = 0.116$ for comparison among tigers and leopards/far-eastern wildcats). Total percentage of animals seropositive to pseudorabies was 13%.

Antibodies to CDV were detected in blood serum from three Amur tigers (including one young animal less than 1.5 years old). One of these males was captured and placed in captivity in 2010 (due to inadequate behavior and loss of fear towards humans). It was kept in captivity for almost a month with medical care but ultimately died. Three consecutive blood samplings on days 1, 17, and 23 showed a high and slightly increasing level of antibodies to CDV. Two other tigers were still alive 3 years after sampling.

Antibodies to CDV were also detected in one far-eastern leopard: one young female had a high titer of antibodies to CDV. No individuals seropositive to CDV were found among far-eastern wildcats and lynxes. The interspecies differences in serum prevalence to CDV were not significant for all pairs of species. In total, antibodies to CDV were found in 4% of cat samples ($n = 44$).

Antibodies to influenza A virus were detected in one Eurasian lynx and in one tiger. We also found a male far-eastern wildcat to be seropositive to influenza A virus. There were no interspecies differences in seroprevalence to the influenza A virus. For all cats, seroprevalence to this pathogen was 7% ($n = 44$).

FHV and FIV both tested seropositive with one animal each (2% of all cats); in both cases – in Amur tigers (the male to FHV and the female to FIV). We found two animals seropositive to FeLV: one male of far-eastern leopard (13%) (this male probably died 1.5 years after it was sampled in August of 2011) and one male of far-eastern wildcat ($n = 11$).

Altogether, 16 (36%) individuals (six wildcats, five leopards, three lynxes, and two tigers) did not have antibodies to any of the tested pathogens. Another 17 (39%) animals (five wildcats, one leopard, one lynx, and 10 tigers) had antibodies to one pathogen. Five animals (11%) (one leopard and four tigers) had antibodies to three pathogens and another six (14%) (four wildcats, one leopard, and one tiger) to two pathogens. Most commonly (seven cases), antibodies to FPV and FCV were discovered together during “co-infections” (co-occurrence of antibodies), and in three cases, we detected antibodies to FPV and pseudorabies virus in the same individuals.

Discussion

We detected the presence of eight viral pathogens in felines at South Primorie. We did not find animals that were seropositive to FCoV. These results are very different from the previous study on tigers from the northern region where 43% of tigers ($n = 44$) were seropositive to FCoV, a value that greatly exceeds the positive one in our study (Goodrich et al. 2012a). The same authors found that four out of 10 tested leopards were also seropositive to FCoV (Goodrich et al. 2012b). These differences may be partially explained by the different methods used for detection of antibodies to FCoV. Our tests also allowed us to find FCoV antibodies in 21% of feral cats at the South Primorie (Goncharuk et al. 2012), but we did not detect seropositive animals among the wild cats. The tests used in this study provide a very reliable estimation of the presence of antibodies to FCoV (Addie et al. 2014). Undoubtedly, such a discrepancy in results of two tiger studies proves the necessity of an additional study of serum samples using different methods.

FPV seems to be very widely distributed at the Russian Far East. Antibodies to FPV were detected in 45% of felines’ serum samples and in 65% of tiger samples. In the central part of the range inhabited by the Amur tiger, 64% of these animals were seropositive to FPV (Goodrich et al. 2012a). The percentage of seropositive tigers there increased with age, and all animals older than 7 years were seropositive to FPV (Goodrich et al. 2012a). Out of the leopards that we caught, three were seropositive to FPV. Earlier, six out of 10 leopards were described to be seropositive to FPV (Goodrich et al. 2012b), which in conjunction with our study shows that 50% of all tested animals were seropositive ($n = 18$). Such a wide distribution of the pathogen may be related to the high resistance of this pathogen to environmental conditions (Wasier et al. 2009). FPV may be very dangerous for wild cats, especially for the survival of their cubs (Stahl and Vandel 1999; Naidenko 2005), which may represent a risk for the stability of wild cat populations at the Russian Far East. However, the high percentage of seropositive animals allows us to suggest that this pathogen did not cause high mortality in tigers, leopards, and far-eastern wildcats. In Eurasian lynx, an absence of seropositive individuals may be related to the high mortality rate of cubs infected with FPV in this species (our data from Tchernogolovka breeding center (Naidenko 2005)) and may be the reason why no antibodies to this virus were found in four wild adult lynxes. For lynxes in this feline community to have never come into contact with this widely distributed virus, which is extremely resistant to weather conditions, is highly improbable. The high mortality rate caused by this virus was also suggested as the explanation for interspecies differences in seroprevalence to this pathogen in wild Pallas’ cat and domestic cat at the Daurskii Reserve (Pavlova et al. 2015).

Antibodies to FCV were detected in Amur tigers and far-eastern leopards for the first time. Only four leopards were tested earlier (Goodrich et al. 2012b) and all of them were seronegative to FCV. In our study, the percentage of felines seropositive to FCV was 22%. Seropositive animals were mainly detected among the far-eastern wildcats. This virus was considered as the cause of death of a captive tiger cub (5 months old, 2013, Tiger’s Rehabilitation Center, Alekseevka (our data)); however, nothing is known about wild populations. The effect of the virus on animal survival was previously estimated only for feral cats (Turnquist and Ostlund 1997; Ossiboff et al. 2007). The antibodies to FCV were detected quite rarely in wild cat populations and never in the area and the species that were subject to the current study. However, the previous data suggest that FCV is widely distributed in wild cats over the world. FCoV antibodies were detected in mountain lions (Puma concolor (Linnaeus, 1771)), ocelots (Leopardus pardalis (Linnaeus, 1758)), oncillas (Leopardus tigrinus (Schreber, 1775)), and different lion populations (the percentage of seropositive animals varied from 0% (Ngorongoro) to 82% (Serengeti)) (Hofmann-Lehmân et al. 1996; Filoni et al. 2006). In feral cats in different parts of Russia, sero-
prevalence may be as high as 62% and correlates positively with population density (Pavlova et al. 2015). In wild cats at the Russian Far East as well as in Southeast Siberia (Pavlova et al. 2015), the level of seroprevalence of FCV is much lower.

CDV has been proven to cause death of Amur tigers (Quigley et al. 2010; Seimon et al. 2013; Gilbert et al. 2015). In 2010, the occurrence of a CDV outbreak was suggested in an Amur tiger population (Seimon et al. 2013). Fifteen percent of tigers were seropositive to CDV in the central part of the Amur tiger range, including the animals that died of CDV infection soon after capture and individuals that survived for years after it (Goodrich et al. 2012a). In our study in South Primorie, the percentage of samples that were seropositive to CDV was 18% for Amur tigers (similar to the data on the central part of the Amur tiger range) and 9% for all felines. One of the three tigers positive for the virus died in captivity, and two others survived for at least 3 years after the sampling. Earlier, two out of 10 tested leopards were found to be seropositive to CDV (Goodrich et al. 2012b), and including our data (one out of eight), the total percentage of leopards seropositive to CDV was approximately the same as in tigers (13% of 18 tested individuals). The low percentage of positivity to CDV in other wild carnivores was described for the south of the Russian Far East earlier (Goncharuk et al. 2012). The high percentage of individuals seropositive to CDV in feral and domestic dogs at the center (58%) (Goodrich et al. 2012a) and at the south (52%) of the tiger’s range (Goncharuk et al. 2012) suggested that they are the main carrier of CDV at the Russian Far East. However, the later study indicated that the virus may also circulate in the wild carnivore community per se (Gilbert et al. 2015). The high mortality rate in CDV-infected carnivores may decrease the percentage of seropositive animals significantly. We did not find animals seropositive to CDV among far-eastern wildcats and lynxes. We assume that far-eastern wildcats are insusceptible to CDV infection similarly to domestic cats (Ohishi et al. 2014). So far, CDV infection has never been described for small cats (genera Felis or in cats of the same size) but CDV has been described previously for lynxes (LYNX CANADENSIS Kerr, 1792 and Lynx Rufus (Schreber, 1777)) (Dausust et al. 2009). CDV is a very dangerous pathogen for large cats, but based on our data, we may suggest that CDV does not have 100% lethality in tigers and leopards. Consequences of infection as well as most probable transmission vectors of CDV at the Russian Far East should be studied intensively for conservation of wild felines.

The influenza A virus can be spread by cats and may often be dangerous for wild cats (Keawcharoen et al. 2004). At the Russian Far East, antibodies to the influenza A virus were found in three individuals (Eurasian lynx, tiger, and far-eastern wildcat). So far, this virus (or antibodies) has never been described for far-eastern leopards. However, the presence of the antibodies to influenza A virus in wild cat populations (that means contact with the pathogen) allows us to consider this virus as potentially dangerous for the big cats in Primorsky krai. Based on the feeding habitats of these feline species, we would assume that the probability of contact with this virus should be higher for smaller cats (lynx and especially far-eastern wildcat), which prey on birds quite often (Yudin 2015).

Animals at Russian Far East were never tested for pseudorabies virus/antibodies before. Five Amur tigers had antibodies to the pseudorabies virus and were tracked after blood sampling for at least 6 months with satellite collars. Usually, wild boars are the main carriers of the pseudorabies virus in the wild (Lari et al. 2006; Vengust et al. 2006; Wilson et al. 2009), and they are the main prey of the Amur tiger (Yudakov and Nikolaev 2012; Kerley et al. 2015). For leopards, wild boars are not the main prey (Pikunov and Korkishko 1992), and Eurasian lynxes and far-eastern wildcats eat wild boar meat mainly as scavengers (but this kind of behavior is not typical for these species). No antibodies to the pseudorabies virus were detected in these three species; however, they were found in two badgers in Ussuriskii Reserve in 2009 that died soon after blood sampling. One of them had clinical signs of pseudorabies (raw scratches on the muzzle), and both of them had antibodies to the influenza A virus, blood parasites (Babesia sp. (Davidova et al. 2010)), and a high number of intestinal parasites (Konyaev et al. 2011). These data points coincide with the epizooty and high mortality rates of badgers and raccoon dogs in Ussuriskii Reserve in the spring of 2009 (Davidova et al. 2010). These small carnivores may be very important carriers of pseudorabies to the large cats (being their common prey) (Pikunov and Korkishko 1992; Kerley et al. 2015) and the far-eastern wildcat (who uses the same dens as badgers and raccoon dogs) (Yudin 2015). Recent findings (Masot et al. 2017) showed that pseudorabies may be lethal for the wild cats, but the probability of this is still unclear. Undoubtedly, research in the wild is needed to answer the questions of how many mammalian species may be susceptible to this virus at the Russian Far East and how lethal this virus is for different host species.

At South Primorie, we also detected the presence of antibodies to FIV in Amur tigers and FeLV antigen in leopards and far-eastern wildcats. In the central area of the tigers’ distribution, more than 40 tiger serum samples were tested and antibodies to these viruses were not detected (Goodrich et al. 2012a) as well as in 10 leopards that were analyzed earlier (Goodrich et al. 2012b). Among 34 feral cats tested in South Primorie, only one was seropositive to FIV (Goncharuk et al. 2012). Based on our results, both of these viruses are present in wild cat populations at South Primorie and may be dangerous for their survival as was described for Iberian lynx (LYNX PARDINUS (Temminck, 1827)) (Meli et al. 2009). It is necessary to note that the FeLV-infected resident leopard disappeared (despite intensive camera-trapping) 1.5 year after blood sampling. His collar malfunctioned and we were unable to track this animal.

These data were collected over the nine years (2008–2016) with possible different occurrence of pathogens. Some epizootic events occurred over this period. First, epizooty of small carnivores occurred in Ussuriskii Reserve in 2009 (Davidova et al. 2010). This study did not find any correlations of diseases in cats with this event. In total, small sample size and more or less equal distribution of capturing success (one to four animals of each species per year) may decrease the chance of ascertaining the effect of epizooty on felines populations. Moreover, our knowledge of epizooties in carnivores or other mammals in this period is limited. However, it seems that we observed the consequences of epizooty. It was described in relation to the 2010 CDV outbreak in the tiger population at the Russian Far East (Seimon et al. 2013). All three seropositive tigers to CDV were captured in October 2009 – May 2010, and all other tigers captured in other periods were seronegative. The main outbreak was described for areas farther north (Sikhote-Alinskii Reserve) (Seimon et al. 2013), which correlated with the period in which it occurred at the South of the Amur tiger range. In 2015, a far-eastern leopard seropositive to CDV was the only animal that was discovered to have CDV antibodies. Possibly, the 2010 CDV outbreak in Amur tigers was related to the genetically modified virus strain, similar to what was described for Serengeti epizooty in lions and hyenas (Nikolin et al. 2017). Additional research is needed on the genetic specifics of CDV to prove this hypothesis.

In summary, we detected the presence of eight (out of nine tested) virus pathogens in felids at South Primorie. For the first time, antibodies to influenza A virus, pseudorabies virus, FIV, and FeLV, which should be considered as a potential threat to some felines, were found in wild cats. Although the data were collected over a few years (due to the difficulties and restrictions of capturing) and seroprevalence could be affected by epizooties of different viruses in these years, these results allow us to conclude that these eight viruses are indeed present in the wild at the Russian Far East. Seroprevalence to most pathogens was significantly higher in wild cats at Southern Primorie than, for example, in Pallas’ cat inhabiting steppes of the Russian Far East (Naidenko et al. 2017).
CDV may result in death of wild large felids at the Russian Far East. Contact with pathogens, the main carrier of the virus. Thirdly, antibodies were present only in tigers, which relates to their trophic relations in host serum may depend on the type of virus and the type of contact with pathogens. Even if the animal was infected by both viruses, it still may have had contact with these pathogens at a different time. To understand co-infections by these pathogens, we need to obtain and analyze viruses per se.

We tested the presence of antibodies to nine pathogens in felids using a non-species-specific test. They were validated previously on domestic cats (FCoV, FPV, FHV, FCV, FIV, and FeLV kits) and a non-species-specific test. They were validated previously on domestic cats (FCoV, FPV, FHV, FCV, FIV, and FeLV kits) and a non-species-specific test. They were validated previously on domestic cats (FCoV, FPV, FHV, FCV, FIV, and FeLV kits) and a non-species-specific test. They were validated previously on domestic cats (FCoV, FPV, FHV, FCV, FIV, and FeLV kits) and a non-species-specific test.


