

Infection with SARS-CoV-2 lineage B.1.1.7 in Three Malayan Tigers at the Virginia Zoological Park

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Short Report

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Abstract

We report three cases of SARS-CoV-2 lineage B.1.1.7 infection in Malayan tigers at the Virginia Zoo. All three animals exhibited respiratory signs. These findings show the mutations in the B.1.1.7 lineage did not affect the susceptibility of tigers to SARS-CoV-2.

Main Text

On April 4, 2021, a five-year-old male Malayan tiger (*Panthera tigris jacksoni*) residing at the Virginia Zoo in Norfolk, Virginia, USA began exhibiting signs of illness including coughing, intermittent upper respiratory sounds consistent with congestion, hyporexia, lethargy, mucoid nasal discharge, and labored breathing. A second Malayan tiger, also a five-year-old male, experienced cough, clear nasal discharge, hyporexia, and labored breathing beginning April 7. The zoo's third tiger, a 10-year-old male, developed a cough on April 10 and clear nasal discharge was observed on April 13. Clinical signs in all three tigers resolved by April 15.

Nasal swabs and fecal samples were collected from the two tigers on April 9 and from the ten year old on April 13 and submitted to the Cornell Animal Health Diagnostic Center for testing. Samples were tested for common feline respiratory pathogens including *Bordetella* sp., *Chlamydia felis*, *Mycoplasma cynos*, *Mycoplasma felis*, *Streptococcus equi*. ssp *zooepidemicus*, Influenza virus, pneumovirus, feline calicivirus and feline herpesvirus, all of which were not detected. All samples tested positive for SARS-CoV-2 using the EZ-SARS-CoV-2 RT-PCR assay and SARS-CoV-2 was isolated from respiratory and fecal specimens from the first tiger. Confirmatory SARS-CoV-2 testing performed by U.S. Department of Agriculture National Veterinary Services Laboratories was positive as well. Screening of the samples with the ThermoFisher COVID-19 TaqPath kit revealed a spike gene dropout, with only the N and Orf1ab gene targets being detected, suggesting potential infection with a B.1.1.7 variant.

Whole genome sequencing of all samples was run using an Oxford Nanopore Technologies MinION as previously described(1). Reads were assembled using the ARTIC ncov-2019 protocol using Medaka for variant calling (<https://artic.network/ncov-2019/ncov2019-bioinformatics-sop.html>). Near-complete (29,702–29,710 bp) assemblies were obtained from the nasal swabs of all three tigers. Sequences have been submitted to GenBank under accessions MZ305031, MZ305032, and MZ305033. High quality assemblies were not obtained from fecal samples. Genomes from the respiratory specimens were identified as belonging to lineage B.1.1.7 using Pangolin v. 2.4.2 (github.com/cov-lineages/pangolin). Nextstrain was used to conduct a phylogenetic analysis of the nasal swab sequences in relation to other B.1.1.7 sequences downloaded from GISAID (accessed April 15, 2021)(2,3). Sequences from the tigers were identical to one another with the exception of one manually corrected homopolymer repeat error and fell into a clade defined by a C4900T mutation containing other samples collected primarily in the United States. The tiger sequences were differentiated from the rest of the clade by one unique SNP in the spike gene (G23236T, Spike K558N). The vdb tool was used to query the GISAID database (accessed May 6, 2021) for other occurrences of this mutation(4). While it was found in 1210 isolates including 506

belonging to lineage B.1.1.7, none shared the C4900T mutation defining the grandparent node of the tiger sequences (Figure).

The source of infection is unknown. While the zoo has been open to the public, the setup of the tiger exhibit makes transmission from a visitor unlikely as all areas of the tiger exhibit are either enclosed by glass or are separated from the public by distances of at least 9 meters. Given the potential for close contact with the infected animals, the most plausible explanation is that one or more of the tigers acquired the virus from a zoo employee, though no employees had a positive SARS-CoV-2 test nor presented symptoms consistent with COVID-19 in the four weeks prior to symptom onset in the tigers. It is possible that all three tigers were infected by a person or that transmission occurred between the tigers. Two of the tigers were kept in the same enclosure and while the third had no direct contact with the others all three rotated through common spaces in their enclosures.

After the tiger infections were identified, four additional animals from the zoo were tested for SARS-CoV-2: one lion (*Panthera leo*) with lethargy and hyporexia starting approximately one week after the diagnosis in the tigers, a second lion that was asymptomatic but tested because of age and proximity to the first lion, and two degus (*Octodon degus*) that died in late March and had interstitial pneumonia found on necropsy. These four animals were negative for SARS-CoV-2.

This report underscores the susceptibility of felids to SARS-CoV-2. Infections have previously been identified in captive lions, tigers, snow leopards, and pumas, as well as in domestic cats. Other non-human animal species including gorillas, dogs, minks, ferrets have also acquired SARS-CoV-2 and additional species have been shown to be susceptible in laboratory settings(5–7). B.1.1.7 lineage infections have been reported in several domestic cats and dogs in the United Kingdom and United States, suggesting that the Spike protein mutations that characterize this lineage have not constrained its host range(8,9). Monitoring for SARS-CoV-2 infection in animals is critical to understanding potential host range of the pathogen, particularly new variants emerge and proliferate.

Declarations

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Conflicts of Interest

The authors declare no conflict of interest.

Author Bio

Dr. Mitchell is a research associate in the Department of Population Medicine and Diagnostic Sciences at Cornell University. His primary research interest is molecular epidemiology of infectious diseases.

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Figures

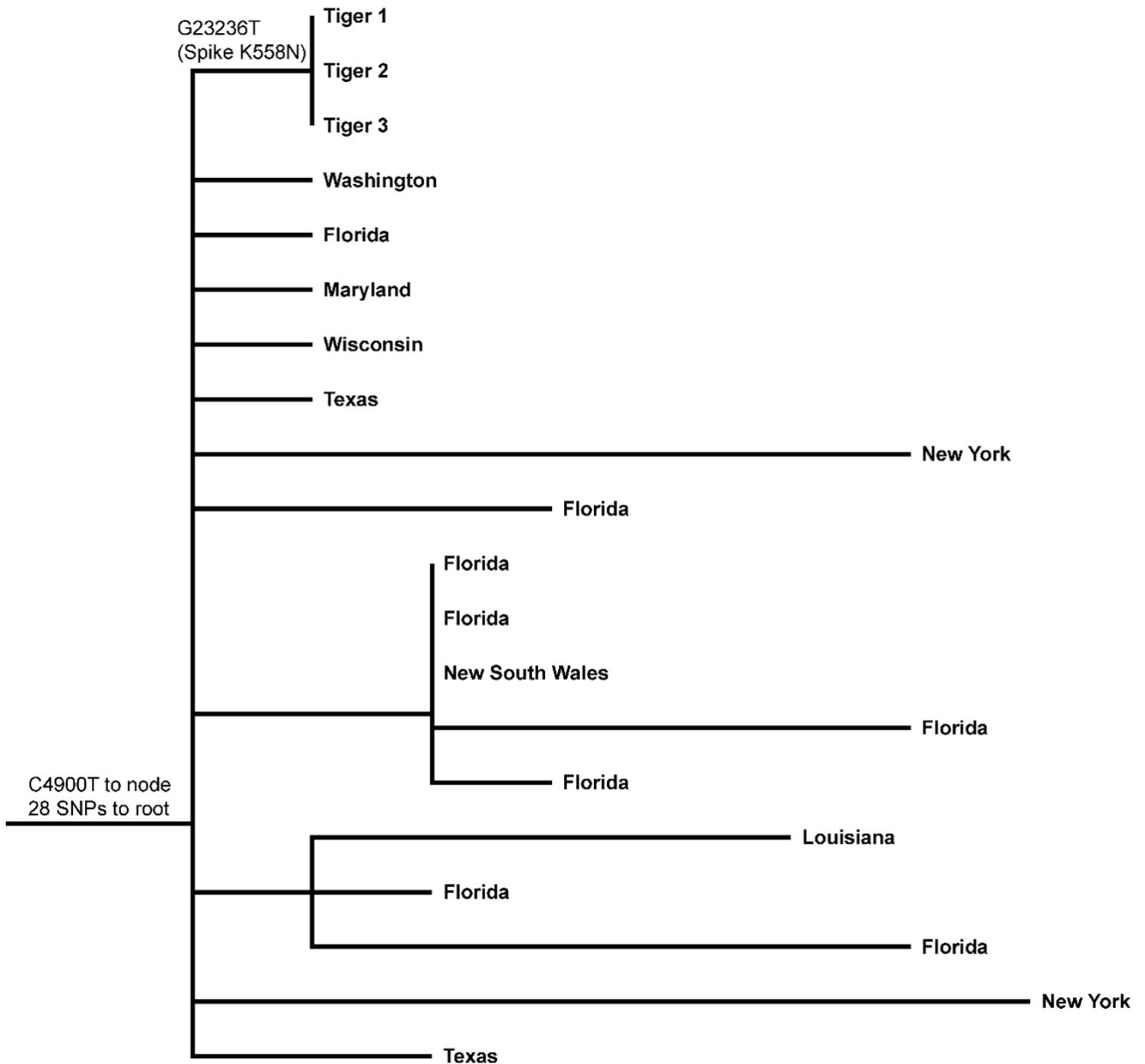


Figure 1

Subset of phylogenetic tree showing parent (G23236T) and grandparent (C4900T) nodes of the tiger sequences, with tips labeled as United States or Australian state of origin. Tiger samples are numbered in order of symptom onset.

Supplementary Files

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- IFAtiger.tiff