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GENOME SEQUENCES

Proposed New Strain of Canine *Kobuvirus* from Fecal Samples of Brazilian Domestic Dogs

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ABSTRACT A proposed new strain of canine *Kobuvirus* was identified in fecal samples of domestic dogs from a rural community located in the municipality of Peixe-Boi, Pará, Brazil. The nucleotide identity was 92.3% similar to other representatives of the family *Picornaviridae*, genus *Kobuvirus*, and species *Aichivirus A*, which suggests that this is possibly a new strain within this species.

Kobuvirus (KoV) belongs to the *Picornaviridae* family, which includes pathogens that infect humans and animals. Since the discovery of the first KoV in a patient with gastroenteritis in March 1989 in Japan (1), other strains have been identified as members of the genus *Kobuvirus* from a variety of domestic and wild animal species, including canine, feline, murine, avian, ovine, bovine, musteline, caprine, and porcine species and wild boars in several regions of the world (2). KoVs are icosahedral and nonenveloped particles which present a genomic organization similar to that of other members of the *Picornaviridae* family and have a linear and positive-sense single-stranded RNA (ssRNA) genome of 8.2 to 8.4 kb, containing a large open reading frame (ORF) coding a single-polyprotein precursor and a 5′ and a 3′ untranslated region (UTR). The polyprotein generates three structural proteins and eight nonstructural proteins (3). In October 2016, 35 fecal samples of nondiarrheic dogs, aged between 5 months and 8 years, were collected at the same time from a rural community located in the municipality of Peixe-Boi, Pará, Brazil.

A cDNA library was prepared and sequenced on an Illumina MiniSeq platform with the methodology described in the Nextera XT DNA library preparation kit (4) with 150-bp paired-end reads. The genome was assembled using a hybrid methodology of *de novo* assembly and reference mapping with the Minia program (kmer size, 32; abndance-min, 3; abundance-max, 500; nb-cores, 8) (5) and the Geneious (default parameters) version 8.1.9 (6) program, respectively. The taxonomic annotation was performed with Kraken software (default parameters) (7). The sequencing produced 3,619,638 reads, and 22,224 contigs were generated, 52 of which were related to the *Picornaviridae* family. The resulting sequence was subsequently characterized as *Canine Kobuvirus* 26 (CaKoV-26) after sequence editing, which was performed on ambiguous bases, and the unitig was inspected and compared with the reference sequence. The genome showed a final length of 8,312 bp. The assembly with reference mapping showed that 390 reads generated a single unitig related to the canine *Kobuvirus* CH-1 strain, species *Aichivirus A* (GenBank accession no. JQ911763), which was the closest sequence using Kraken, presenting 92.3% nucleotide identity with a new strain, CaKov-26. In the P1, P2, and P3 polyprotein regions, the nucleotide identities were 89.6%, 95.9%, and 94.9%, and the amino acid identities were 94%, 99.8%, and 98.7%, respectively. The average coverage of reads was 7.1×, and the GC content was 58.1%. The

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average similarity between the different KoVs was 92% to 98%, and the CaKoV-26 strain showed a common identity that is related to these viruses. The ORF search to define the polyprotein region, domains, and subdomains was annotated by comparison with 59 other *Picornaviridae* genome sequences.

Several studies in Brazil have demonstrated the circulation of KoV strains in different host species and geographical locations (8–14), but herein, we describe the first complete genome sequence of the new canine *Kobuvirus* strain in nondiarrheic dogs and highlight the importance of KoV in the context of infections. Since there are few studies about this virus, we aim to contribute to a better understanding of its epidemiological, pathogenic, and zoonotic aspects.

Data availability. The complete genome sequence of canine *Kobuvirus* strain CaKoV-26 was deposited in GenBank under the accession no. MH747478.

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