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PORCINE VIRUSES IN THE WILD BOAR POPULATION OF THE CENTRAL FEDERAL DISTRICT OF RUSSIA^{*}

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Abstract

Porcine viral diseases cause significant harm to the pig industry. Wild boars can be a natural reservoir of domestic pig's pathogens. Thus, monitoring studies directed on their infectious status help veterinary specialists to determine the current epizootic situation in the country. The study of the prevalence of porcine viruses in the wild boar population in the Central Federal District of Russia is necessary due to the presence of intensive pig farming.

Between 2021 and 2023, samples were collected from 79 hunter-harvested wild boars in three regions of Russia: 30 from Moscow, 24 from Tver', and 25 from Belgorod Regions. From each hunted animal, three different sample types were taken: bronchial lymph node, lung, and spleen. Collected samples were transported to the laboratory under freezing conditions. From each organ sample, a piece of tissue was taken and homogenized in saline solution, the prepared samples were aliquoted in 1.5mL Eppendorf tubes and stored at -70° C. The total DNA was extracted from the supernatant of organ suspension using the commercial kit "RIBO-prep" (FBIS Central Research Institute of Epidemiology of Rospotrebnadzor, Moscow, Russia) following the manufacturer's instructions.

The samples were tested for the presence of porcine reproductive and respiratory syndrome virus (PRRSV), classical swine fever virus (CSFV), porcine circoviruses 2-4 (PCV-2-4), and porcine parvoviruses 1-7 (PPV-1-7). To detect genomes of PRRSV, CSFV, PCV-2, and PPV-1, real-time PCR (qPCR) was conducted using a commercial PCR test kit (Vetbiochem, Moscow, Russia) according to the manufacturer's instructions. For PCV-3, PCV-4, PPV-2-7 detection, primers and probes were used from the previous studies [1–6]. Samples with the best Ct-value in qPCR or the best quality during electrophoresis were subjected to Sanger genome sequencing and genotyping for further phylogenetic analysis. DNA sequence chromatograms were analyzed and assembled into final consensus using SeqMan Lasergene 11.1.0. (DNASTAR, Madison, WI, United States). The phylogenetic analysis was performed using MEGA 7.0 software.

Results of this study are presented in Table 1 (see table). Porcine pathogens PCV-2 and PPV-1 were detected in 63.3 and 25.3 % of animals, respectively. PCV-3, which pathogenicity has not been proven yet, was found in 41.8 % of wild boars. PPV-2-7 were also detected in 30.4, 53.2, 1.3, 2.5, 7.6, 59.5 %, respectively. Genomes of the PRRSV, CSFV and PCV-4 were not found in any of the studied samples.

| | Infected wild boars, animals | | | Total number of infected wild boars | |
|-------|------------------------------|---------------------|------------------------|-------------------------------------|------|
| | Moscow Region | Tver' Region | Belgorod Region | Animals | % |
| PCV-2 | 19 | 12 | 19 | 50 | 63,3 |
| PCV-3 | 7 | 13 | 13 | 33 | 41,8 |
| PPV-1 | 6 | 6 | 8 | 20 | 25,3 |
| PPV-2 | 5 | 11 | 8 | 24 | 30,4 |
| PPV-3 | 11 | 15 | 16 | 42 | 53,2 |
| PPV-4 | 0 | 0 | 1 | 1 | 1,3 |
| PPV-5 | 1 | 1 | 0 | 2 | 2,5 |
| PPV-6 | 2 | 0 | 4 | 6 | 7,6 |
| PPV-7 | 21 | 11 | 15 | 47 | 59,5 |

Distribution of porcine viruses in wild boars in the regions of the Russian Federation

The phylogenetic analysis was performed for PCV-2, PCV-3, and PPV-1. We sequenced 23 genomes of PCV-2: 15 from Moscow, seven from Tver', and one from Belgorod Regions, respectively. Phylogenetic analysis for PCV-2 was performed by ORF2 (*Cap* gene). Finally, 21/23 isolates belonged to PCV-2d genotype, two isolates from Moscow region belonged to PCV-2b. For PCV-3, four complete genomes from Moscow and two from Tver' regions were sequenced. All

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sequenced isolates belonged to PCV-3a genotype. For PPV-1 phylogenetic analysis, ORF2 (VP1/VP2) were used. We sequenced five isolates from Moscow, one from Tver', and four from Belgorod Regions. According to the result of phylogenetic analysis, PPV-1 genotypes of both Asian and European lineages were identified among wild boars.

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