Establishment of Two Novel Phylogenetic Lineages of PRRSV-2 from Canada

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Introduction and Objectives: In Canada, Porcine Reproductive and Respiratory Syndrome Virus 2 (PRRSV-2) causes annual losses of approximately USD 150 million, and roughly 6.4 million live pigs are exported annually to the United States. Canada has been suggested as a historical source of certain PRRSV-2 sub-lineages introduced into the U.S.; however, these conclusions are based on outdated studies or analyses with a limited number of contemporary sequences. As a result, our current understanding of PRRSV-2 diversity in Canada remains incomplete. Here, we characterize the diversity of PRRSV-2 in Canada and place it within the context of global classification.

Materials and Methods: We analyzed a total of 3,573 PRRSV-2 ORF5 sequences from Canada, collected across five provinces between 2000 and 2024. Sequences were classified into sub-lineages using PRRS-Loom, and genetic distances among detected sub-lineages were estimated using SDTv1.2.

Results and Discussion: We identified 13 PRRSV-2 sub-lineages circulating in Canada (1A, 1B, 1C, 1E, 1F, 1H, 1I, 1J, 5A, 7, 8A, 8C and 9A), as well as four highly divergent monophyletic clades with strong bootstrap support (>99%) that did not correspond to any previously described sub-lineage. Sequence comparisons indicated that these clades exhibit more than 9.5% genetic dissimilarity relative to known sub-lineages. All four monophyletic clades are most closely related to Lineage 1. Two of these clades have not been detected since 2021 and appear at low frequency in our dataset, whereas the other two have remained in circulation since the early 2000s, each representing more than 10% of wild-type sequences in our dataset. Based on their epidemiological relevance, we propose two new sub-lineages, 1K and 1L. Our data suggest that sub-lineage 1K is also present in the United States, but circulating at low frequency, whereas sub-lineage 1L appears to be restricted to Canada

Conclusions: Our results expand the current understanding of PRRSV-2 genetic diversity. Here, we establish two novel sub-lineages within Lineage 1. Our findings further reinforce that the global diversity of PRRSV-2 remains underexplored, and that additional, yet undescribed sub-lineages may be circulating in other regions. Furthermore, these results highlight the need for a universal nomenclature and classification system to improve communication and support the implementation of more effective biosecurity measures.

The full paper published in Pathogens can be accessed at: https://doi.org/10.3390/pathogens15040346

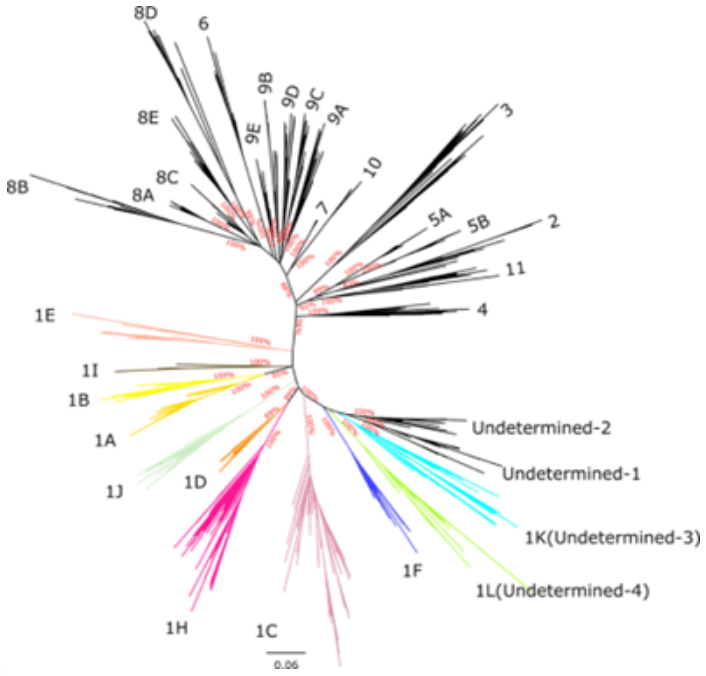

Figure 1. Unrooted maximum-likelihood phylogenetic tree based on ORF5 anchor sequences representing all sub-lineages globally, including newly identified Canadian sub-lineages and undetermined clades 1 and 2. The tree was constructed using RAxML-NG, employed the GTR+I+G4 substitution model. Numbers on the branches indicate branch support values for the basal nodes of each sub-lineage. The scale bar at the bottom represents the number of substitutions per site.