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Publication date:
2012

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Citation (APA):

Larsen, L. E., Hjulsager, C. K., Kvisgaard, L. K., Wellenberg, G. J., Botti, S., Frossard, J. P., ... Nauwynck, H. J. (2012). Diversity of type i porcine reproductive and respiratory syndrome virus (PRRSV) in europe: A PORRSCON study. Abstract from 22nd International Pig Veterinary Society Congress, Jeju, Korea, Republic of.

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DIVERSITY OF TYPE I PORCINE REPRODUCTIVE AND RESPIRATORY SYNDROME VIRUS (PRRSV) IN EUROPE: A PORRSCON STUDY

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Introduction

Since its discovery in the late 1980s, PRRSV has emerged to be one of the most prevalent infectious diseases of pigs worldwide. In the US alone, the annual costs of PRRSV have been estimated at \$560 million¹. Recently, studies have shown that some European countries such as Lithuania, Latvia, Belarus and Russia harbor exceptionally diverse EU-genotype PRRSV strains², but updated PRRSV sequence data from the rest of Europe are very sparse. Some PRRSV strains have been shown to be especially virulent, emphasizing the need for continuous and global monitoring of PRRSV diversity. The aim of the present study was to compile and assess information on the diversity of PRRSV circulating in Europe in recent years.

Materials and Methods

PRRSV viruses collected through active and/or passive surveillance programs in Denmark, Belgium, UK, Italy, Spain, Poland, The Netherlands and France were included in the analysis (see table 1 for number of isolates supplied from each country and the year of collection). RNA was extracted directly from tissue or serum from infected animals or fetuses or from virus isolates passaged in MARC-145 cells or Pulmonary Alveolar Macrophages (PAMs). PCR amplification and DNA sequencing of ORF5 of the PRRSV genome was performed using assays typically used in the different countries. Consensus sequences were generated in each lab and the sequences sent in Fasta format to the lead author for phylogenetic analysis. Alignment and Neighbour-Joining trees with 1000 bootstrap replicates were performed with CLC DNA Workbench v.6.0.1 (CLC BIO, Aarhus, Denmark). Trees were drawn with FigTree v1.3.1 (<http://tree.bio.ed.ac.uk/software/figtree/>)

Results and Discussion

ORF5 from a total of 101 PRRSV samples from eight European countries were included in the analysis (table 1). Apart from the Italian and French isolates, all viruses were collected in 2010/11. Overall, the identity between isolates was 80-100% at the genetic level. The national diversity varied among the countries (Table 1), but this may merely reflect the differences in time and interval of

collection. The phylogenetic analysis performed showed that all of the isolates belonged to genotype 1. In general, isolates from within a country clustered together in the phylogenetic tree, but several countries had isolates that clustered in distinct clusters. Interestingly, some clusters contained nearly identical viruses isolated in different countries. These findings indicate that PRRSV (recently) crossed borders via trade of semen or live animals, however, the high number of national clusters also indicate that in general the national PRRSV populations in the different European countries are separated.

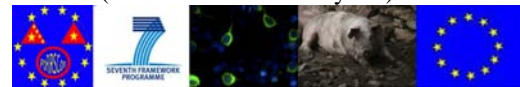
In four different countries, ORF5 sequences very closely related to the Lelystad vaccine virus were identified. This may indicate either that the vaccine strain had spread from herd to herd, that animals in the herd were vaccinated at the time of sampling or that the Lelystad-like lineage of viruses has persisted in several countries.

Table 1. Range of national diversity of ORF5

Country	Year	# of seq	% nt identity
Belgium	2010	4	84-91
Denmark	2010-2011	11	85-100
France	1992-2005	5	89-97
Italy	2008-2009	19	82-99
The Netherlands	2010-2011	8	82-95
Poland	2010-2011	20	81-99
Spain	2011	10	90-99
UK	2010-2011	24	86-99

Acknowledgements

The research leading to these results has received funding from the European Union Seventh Framework Programme (FP7/2007-2013) under grant agreement n° 245141 (coordinator H. Nauwynck).



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